GSC Biological and Pharmaceutical Sciences
eISSN: 2581-3250 CODEN (USA): GBPSC2
Cross Ref DOI: 10.30574/gscbps
Journal homepage: https://gsconlinepress.com/journals/gscbps/

(RESEARCH ARTICLE)

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In vitro responses of *Dendrobium* sp. plantlets to Gibberellic Acid (GA_3) under PEG 6000-induced drought stress

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GSC Biological and Pharmaceutical Sciences, 2026, 35(01), 157-166

Publication history: Received on 02 March 2026; revised on 12 April 2026; accepted on 14 April 2026

Article DOI: https://doi.org/10.30574/gscbps.2026.35.1.0135

Abstract

Orchids are among the most economically important ornamental plants, with *Dendrobium* sp. widely cultivated due to its high diversity and commercial value. However, drought stress remains a major constraint, affecting plant growth at morphological, physiological, and anatomical levels. The application of gibberellic acid (GA_3) at an appropriate concentration is a potential strategy to improve plantlet performance under stress conditions. This study aimed to determine the optimal GA_3 concentration and to evaluate the morphological, physiological, and anatomical responses of *Dendrobium* sp. plantlets under *In vitro* drought stress conditions. The experiment was arranged in a Completely Randomized Design (CRD) with a single factor consisting of five GA_3 concentrations (0, 1, 2, 3, and 4 mg/L) in VW medium supplemented with Polyethylene Glycol (PEG 6000). Each treatment consisted of five replicates, with one plantlet per culture bottle. Data were analyzed using analysis of variance (ANOVA) at a 5% significance level, followed by Duncan's Multiple Range Test (DMRT). The results showed that GA_3 concentration significantly affected stomatal density, while other parameters showed clear trends without significant differences. The optimal GA_3 concentration was 1 mg/L, which resulted in higher plantlet survival, improved morphological performance, increased chlorophyll content, and higher stomatal density. In contrast, higher GA_3 concentrations (≥2 mg/L) tended to reduce plantlet performance, indicating a dose-dependent inhibitory effect under drought stress conditions. These findings demonstrate that GA_3 plays a dose-dependent role in regulating plantlet responses to drought stress and highlight the importance of optimizing plant growth regulator application in orchid tissue culture systems.

Keywords: *Dendrobium* sp.; Drought Stress; Gibberellic Acid (GA_3); *In vitro*; Polyethylene Glycol (PEG 6000)

1. Introduction

Dendrobium orchids constitute one of the most important genera in horticulture and tissue culture, valued for their aesthetic appeal and substantial commercial potential. *In vitro* culture techniques are extensively utilized for large-scale plantlet propagation and for preserving desirable genetic variants. However, plantlets grown under *In vitro* conditions are highly vulnerable to abiotic stresses, particularly drought stress, which can significantly impair their growth, development, and overall viability.

Growth hormones such as gibberellins (GA_3) are known as key regulators of plant growth and development. In *In vitro* culture systems, GA_3 is commonly applied to stimulate cell division, shoot elongation, and organogenesis [1]. However, the effects of GA_3 under water-deficit conditions remain incompletely understood, particularly in relation to its role in enhancing plantlet tolerance and physiological responses during drought stress.

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Studies in model plants have demonstrated that Gibberellins (GA) signaling pathways are closely associated with drought tolerance. Reduced GA activity, or the regulation of GA through DELLA proteins, can enhance osmotic stress tolerance by decreasing stomatal conductance and maintaining water-use efficiency [2]. In addition, GA_3 interacts with other stress-related hormones such as abscisic acid (ABA) in regulating stomatal closure and controlling water loss [3].

Drought stress also influences the expression of GA biosynthesis genes and the development of stomatal morphology. In tomato, drought conditions reduce the expression of GA-related genes and inhibit cell expansion, which subsequently affects plant growth and morphological adaptation [4]. Thus, the application of exogenous GA_3 may modulate growth, stomatal density, and drought tolerance; however, its specific mechanisms in *Dendrobium* plantlets remain largely unexplored.

Based on this background, this study aims to evaluate the effects of GA_3 on the growth, viability, and stomatal density of *Dendrobium* sp. plantlets under simulated drought stress (PEG 6000) in tissue culture. This study also aims to identify the optimum GA_3 concentration that can support the physiological adaptation of plantlets to water stress.

2. Materials and Methods

2.1. Tools and Materials

The tools and materials used in this study included an autoclave, a laminar airflow cabinet (ESCO), a Bunsen burner, an analytical balance, Erlenmeyer flasks, beakers, measuring cylinders, culture bottles, glass rods, funnels, test tubes, test tube racks, plastic trays, forceps, scalpels, scissors, a mortar and pestle, droppers, micropipettes with sterile tips, cuvettes, a pH meter, and a UV–Vis's spectrophotometer.

The biological material used was *Dendrobium* sp. plantlets. The chemical reagents included 70% and 96% ethanol, sterile distilled water, Vacin and Went (VW) medium, sucrose, agar, potassium hydroxide (KOH), hydrochloric acid (HCl), gibberellic acid (GA_3), label paper, Whatman No. 1 filter paper, tissue paper, aluminum foil, plastic wrap, rubber bands, detergent, and commercial bleach.

2.2. Experimental Design and Data Analysis

This study employed a Completely Randomized Design (CRD) with a single factor, namely GA_3 concentration (0, 1, 2, 3, and 4 mg/L). All media were supplemented with 20% polyethylene glycol (PEG 6000). Each treatment consisted of five replicates, with one plantlet per culture bottle. The observed parameters included the percentage of surviving plantlets, plantlet morphological characteristics, stomatal density, and chlorophyll content.

The experiment was conducted from November to December 2024 in the Tissue Culture Facility, Botany Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Lampung. Data obtained from each variable were analyzed using Analysis of Variance (ANOVA) at a significance level of $\alpha = 5\%$. When significant differences among treatments were detected, the analysis was followed by Duncan's Multiple Range Test (DMRT) at the 5% significance level to compare treatment means.

2.2. Methods

2.2.1. Preparation of Control and Treatment Media

The preparation of the control medium was initiated by preparing all required tools and materials. The components were weighed using an analytical balance. A ready-to-use Vacin and Went (VW) medium (0.344 g/200 mL) was mixed with sucrose (6 g/200 mL) and activated charcoal (0.4 g/200 mL). Sterile distilled water was then added to reach a final volume of approximately 200 mL.

The medium was homogenized using a magnetic stirrer on a hotplate. The solution was heated until completely dissolved, and the pH was adjusted to 5.7 using 1 N HCl (if the medium was too alkaline) or 1 N KOH (if the medium was too acidic).

After pH adjustment, agar (1.4 g/200 mL) was added and heated while stirring until fully dissolved and the solution reached boiling point. Polyethylene glycol (PEG 6000) at a concentration of 20% was then incorporated into the

medium. Prior to addition, PEG 6000 was dissolved in sterile distilled water and filtered using Whatman No. 1 filter paper under aseptic conditions in a laminar airflow (LAF) cabinet.

The medium was dispensed into culture bottles at 20 mL per bottle, sealed with plastic covers and rubber bands, and sterilized in an autoclave at 121 °C and 17.5 psi for 15 minutes. Sterile GA_3 solutions at different concentrations (0, 1, 2, 3, and 4 mg/L) were added after autoclaving while the medium was still warm under aseptic conditions in the LAF cabinet.

After GA_3 supplementation, the culture bottles were re-sealed and incubated at room temperature (±25 °C) for 7 days. Media that showed no signs of contamination during this incubation period were considered suitable for use in the experiment.

2.2.2. Planting of Dendrobium sp*. Plantlets*

All instruments (forceps and scalpels) were sterilized by flaming over a Bunsen burner prior to use. *Dendrobium* sp. plantlets were removed from the original culture bottles, rinsed with sterile distilled water, immersed in a bleach solution for 3 minutes, and subsequently rinsed again with sterile distilled water.

The sterilized plantlets were then inoculated onto the control medium (20% PEG 6000) and treatment media (20% PEG 6000 supplemented with GA_3 at respective concentrations), with one plantlet per culture bottle. All procedures were carried out aseptically inside a laminar airflow (LAF) cabinet to maintain sterility.

The culture bottles were then sealed, wrapped with plastic film, secured with rubber bands, and labeled accordingly. The cultures were incubated on culture racks under appropriate room temperature and light conditions to support plantlet growth.

2.2.3. Survival Percentage of Plantlets

Observations on the percentage of surviving plantlets were conducted weekly for up to four weeks. The survival percentage of *Dendrobium* sp. plantlets was calculated using the following formula [5]:

$$Survival\ Percentage\ (\%) = \frac{Number\ of\ surviving\ plantlets}{Total\ number\ of\ plantlets}\ x\ 100$$

2.2.4. Plantlet Visualization

Plantlet visualization data were expressed as the percentage of green plantlets, indicating healthy growth status. The percentage of green plantlets was calculated using the following formula [5]:

$$Green\ plantlets\ percentage\ (\%) = \frac{Number\ of\ green\ plantlets}{Total\ number\ of\ plantlets}\ x\ 100$$

2.2.5. Chlorophyll Content

Leaves of *Dendrobium* sp. plantlets (0.1 g, without midribs) were ground using a mortar and pestle with the addition of 10 mL of 80% acetone. The resulting filtrate was collected, and 1 mL of the extract was transferred into a cuvette. Absorbance was measured at wavelengths of 646 nm and 663 nm using a UV–Vis's spectrophotometer.

Chlorophyll content was determined according to [6] using the following equations:

Chlorophyll content was calculated using the following formulas:

- Chla=(12.25×λ663) −(2.79×λ646) (mg/L)
- Chlb=(21.50×λ646) −(5.10×λ663) (mg/L)
- Total chlorophyll=(7.15×λ663) +(18.71×λ646) (mg/L)

Where:

- Chla = Chlorophyll a
- Chlb = Chlorophyll b
- λ646 = Absorbance at wavelength 646 nm

- λ663 = Absorbance at wavelength 663 nm

2.2.6. Stomatal Density

Leaves of *Dendrobium* sp. plantlets were cleaned to remove dust and debris. The abaxial (lower) epidermal surface was then coated with a thin layer of transparent nail polish and allowed to dry for approximately 5 minutes. After drying, the coated surface was covered with clear adhesive tape, which was carefully peeled off to obtain an epidermal replica.

The replica was mounted onto a labeled glass slide and observed under a microscope at 100× magnification. Images were captured using an Optilab camera. Observations were conducted using a calibrated Optic B-510-2F microscope with a field of view area of 3.8 mm^2.

Stomatal density was calculated using the following formula adapted from [7]:

$$\text{Stomatal density (stomata/mm}^2) = \frac{\text{Number of stomata}}{\text{Field of view area (mm}^2)}$$

3. Results and discussion

3.1. Survival Percentage and Plantlet Visualization

Observations on the growth of *Dendrobium* sp. orchid plantlets treated with gibberellic acid (GA_3) for four weeks showed differences in survival rates and morphological visualization among treatments. GA_3 is known to exert both positive and negative effects depending on its concentration; at higher concentrations, GA_3 may become toxic and interfere with physiological processes and cellular metabolism, including in *In vitro* culture systems [8,9]. In this study, GA_3 was applied at five different concentrations: 0 mg/L, 1 mg/L, 2 mg/L, 3 mg/L, and 4 mg/L. Each treatment consisted of five replicates, with one plantlet per culture bottle. The survival percentage and visual characteristics of *Dendrobium* sp. plantlets at different GA_3 concentrations are presented in Table 1.

Table 1 Survival Percentage of *Dendrobium* sp. Plantlets Under Different GA_3 Concentrations Under Drought Stress

GA_3 Concentration (mg/L)	Week 1 (%)	Week 2 (%)	Week 3 (%)	Week 4 (%)
0	100	100	100	100
1	100	100	100	100
2	100	100	100	100
3	100	100	100	100
4	100	100	100	80

Based on the results presented in Table 1, treatments with GA_3 concentrations of 0, 1, 2, and 3 mg/L showed a 100% survival rate of *Dendrobium* sp. plantlets up to the fourth week. However, at the GA_3 concentration of 4 mg/L, the survival rate remained at 100% only until the third week and subsequently declined to 80% in the fourth week. This decline is presumably associated with physiological stress in the plantlets due to the accumulation of GA_3 at higher concentrations, which may disrupt metabolic stability, reduce adaptive capacity, and ultimately decrease tissue viability [10,11]. Similar findings have been reported in orchid and other plant tissue cultures, where excessive application of gibberellins can induce morphological abnormalities, reduce vigor, and inhibit normal growth [12,13]. The visualization of *Dendrobium* sp. plantlets after GA_3 treatment at various concentrations under drought stress conditions is presented in Table 2.

Meanwhile, Table 2 shows that although most plantlets maintained a high survival rate, their visual quality tended to decline with increasing GA_3 concentration. In the control treatment (0 mg/L) and at 1 mg/L, plantlets generally remained green up to the third week, but began to exhibit yellowish-green discoloration in the fourth week.

At concentrations of 2 mg/L and above, the decline in visual quality became more pronounced, as indicated by an increasing proportion of plantlets exhibiting chlorosis, discoloration, and other stress-related symptoms. The reduction in green coloration suggests impairment of physiological processes, such as decreased chlorophyll synthesis and hormonal imbalance due to excessive GA_3 exposure, as reported in various *In vitro* plant cultures, including orchids

[12,14]. High concentrations of GA_3 are also known to induce chlorosis and metabolic disturbances, leading to tissue discoloration and reduced plantlet vigor [11,13].

Table 2 Percentage of Green *Dendrobium* sp. Plantlets Under Different GA_3 Concentrations Under Drought Stress

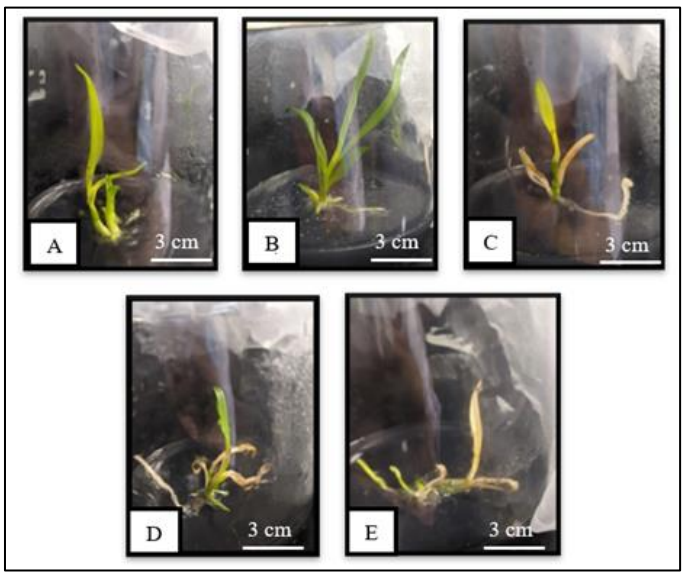
GA_3 Concentration (mg/L)	Week 1 (%)	Week 2 (%)	Week 3 (%)	Week 4 (%)
0	100	100	100	60
1	100	100	80	80
2	100	60	40	40
3	60	20	20	20
4	40	40	0	0

Note: Green plantlets indicate healthy morphological condition, while non-green plantlets include chlorotic or necrotic tissues.

One of the factors influencing the results in Tables 1 and 2 is the adaptation period of plantlets to new growth conditions. *Dendrobium* sp. plantlets generally require a relatively long acclimation period; therefore, changes in environmental conditions may trigger visible morphological responses, such as leaf curling, discoloration, and tissue desiccation. This indicates that excessive GA_3 application may disrupt plantlet metabolism, thereby affect not only tissue coloration and vitality but also potentially reduce long-term survival.

Therefore, determining the optimal concentration of GA_3 is essential to support plantlet growth without inducing excessive physiological stress. These findings are consistent with previous studies indicating that GA_3 at optimal concentrations can enhance plant growth and fulfill physiological requirements, whereas concentrations exceeding the optimum threshold may lead to hormonal imbalance and negatively affect *In vitro* plant growth [11, 15, 16].

The visualization of *Dendrobium* sp. plantlets is presented in Figure 1.



Note: (A) 0 mg/L (control); (B) 1 mg/L; (C) 2 mg/L; (D) 3 mg/L; (E) 4 mg/L.

Figure 1 Morphological Changes of *Dendrobium* sp. Plantlets Under Different GA_3 Concentrations Under Drought Stress Conditions

The visualization results indicate changes in color and morphological condition of plantlets at each GA_3 concentration throughout the observation period. At lower concentrations, plantlets tended to maintain their green coloration and appeared healthy, whereas at higher concentrations, symptoms of physiological stress were observed, characterized by discoloration from green to yellowish and eventually brownish tones.

This phenomenon is consistent with reports indicating that excessive application of GA_3 can induce chlorosis and chlorophyll pigment degradation in *In vitro* cultures [17]. Furthermore, high GA_3 concentrations may increase the

production of reactive oxygen species (ROS), which can damage chloroplast structures and accelerate tissue discoloration under stress conditions [18].

In addition, the use of plant growth regulators (PGRs) at inappropriate concentrations is known to cause abnormal morphology and phytotoxic symptoms in plantlets [19].

3.2. Chlorophyll Content

Chlorophyll content analysis was conducted to evaluate the photosynthetic capacity of *Dendrobium* sp. plantlets, which plays a crucial role in supporting plant growth and vitality. In this study, *Dendrobium* sp. plantlets were cultured on Vacin and Went (VW) medium supplemented with PEG 6000 and treated with various concentrations of GA_3 to assess their physiological responses under drought stress conditions. The mean chlorophyll content of *Dendrobium* sp. plantlets at the fourth week after GA_3 treatment is presented in Table 3.

Table 3 Mean Total Chlorophyll Content of *Dendrobium* sp. Plantlets at Week 4 After GA_3 Treatment

GA_3 Concentration (mg/L)	**Total Chlorophyll Content (Mean ± SE)**
0	2.35 ± 0.886^{a}
1	7.28 ± 1.112^{b}
2	4.04 ± 0.614^{ab}
3	4.68 ± 0.901^{ab}
4	3.84 ± 1.018^{ab}

Note:Values are expressed as mean ± standard error (SE). Different superscript letters indicate significant differences among treatments based on Duncan's Multiple Range Test (DMRT) at $p \leq 0.05$.

Chlorophyll content analysis was conducted to evaluate the photosynthetic capacity of *Dendrobium* sp. plantlets grown on VW medium supplemented with PEG 6000 under different GA_3 concentrations. Total chlorophyll content was measured at week 4 after treatment to assess plantlet physiological responses under drought stress conditions as well as the effect of GA_3 application.

Based on Table 3, total chlorophyll content varied among GA_3 treatments. Although statistical analysis indicated no significant differences among treatments, a clear numerical trend was observed. The highest chlorophyll content was recorded at 1 mg/L GA_3 (7.28), whereas the lowest value was found in the control (0 mg/L) at 2.35. This pattern suggests that plantlets still maintained physiological tolerance to GA_3 application under drought stress conditions.

Overall, chlorophyll content tended to decrease with increasing GA_3 concentrations beyond the optimum level. At 1 mg/L, chlorophyll content was relatively higher and more stable compared to other treatments, indicating that this concentration may represent the optimal level for supporting chloroplast metabolism and maintaining photosynthetic capacity. At optimal concentrations, GA_3 is known to promote cell expansion, maintain chloroplast membrane integrity, and reduce chlorophyll degradation, particularly under abiotic stress conditions.

In contrast, higher GA_3 concentrations—especially at 4 mg/L—resulted in reduced chlorophyll content. This decline is likely associated with supra-optimal hormonal effects, where excessive GA_3 may disrupt endogenous hormonal balance, impair chloroplast structure, and accelerate the degradation of photosynthetic pigments under drought stress. Such responses are commonly linked to increased oxidative stress and metabolic imbalance in plant tissues.

These findings are consistent with previous studies reporting that gibberellins at appropriate concentrations can enhance cell division, elongation, and photosynthetic efficiency. However, excessive application may lead to inhibitory effects on plant physiological processes. Therefore, GA_3 at 1 mg/L can be considered the most effective concentration for maintaining chlorophyll content in *Dendrobium* sp. plantlets, while higher concentrations tend to reduce photosynthetic performance.

3.3. Stomatal Density

Stomatal density is an important parameter reflecting plant responses to drought stress, as it is closely related to transpiration regulation and gas exchange. Lower stomatal density is often associated with improved water-use efficiency (WUE), although stomatal responses are highly plastic depending on stress intensity and plant characteristics

[25,26]. In addition to density, stomatal size and aperture also influence physiological performance under stress conditions [27]. The stomatal density of *Dendrobium* sp. plantlets under different GA_3 concentrations is presented in Table 4.

Table 4 Stomatal Density of *Dendrobium* sp. Plantlets at Different GA_3 Concentrations

GA_3 Concentration (mg/L)	**Stomatal Density (Mean ± SE)**
0	4.50 ± 0.246^{ab}
1	6.16 ± 0.124^{c}
2	4.74 ± 0.325^{b}
3	3.90 ± 0.167^{a}
4	4.42 ± 0.148^{ab}

Note: Values are expressed as mean ± standard error (SE). Different superscript letters indicate significant differences among treatments based on Duncan's Multiple Range Test (DMRT) at $p \leq 0.05$.

The results presented in Table 4 indicate that GA_3 concentration significantly affected stomatal density of *Dendrobium* sp. plantlets. The highest stomatal density was observed at 1 mg/L GA_3 (6.16 ± 0.124), which was significantly different from other treatments, while the lowest value was recorded at 3 mg/L (3.90 ± 0.167).

The increase in stomatal density at 1 mg/L suggests that this concentration may enhance leaf developmental processes, particularly epidermal cell differentiation and stomatal formation. Gibberellins are known to promote cell division and elongation, which can influence stomatal development under controlled conditions.

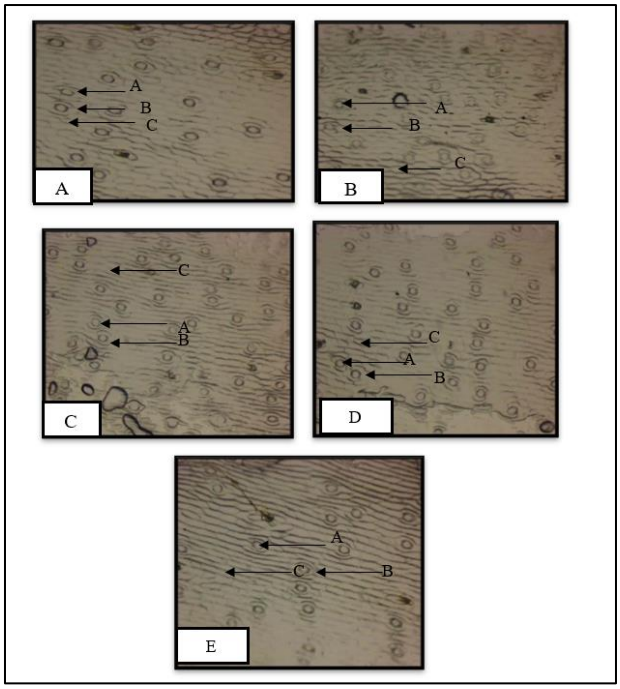
However, at higher GA_3 concentrations (2–4 mg/L), stomatal density decreased, indicating a potential inhibitory effect at supra-optimal levels. This reduction may represent an adaptive response to drought stress conditions induced by PEG 6000, where plants reduce stomatal number to limit water loss through transpiration and improve water-use efficiency (WUE) [25]. Similar responses have been reported under severe stress conditions, where both stomatal density and size may decrease as part of plant survival strategies [26].

The observed pattern supports the dose-dependent role of GA_3, where optimal concentrations stimulate stomatal formation, while higher concentrations exert inhibitory effects [28,29]. Such responses may be associated with hormonal crosstalk and the regulation of the GA–DELLA signaling pathway, which plays a key role in controlling cell differentiation and stomatal development [30].

Interestingly, this trend is consistent with chlorophyll content results, where 1 mg/L GA_3 also produced the highest values, while higher concentrations led to a decline. This indicates that excessive GA_3 may disrupt physiological balance, affecting both stomatal development and photosynthetic performance.

Overall, these findings suggest that GA_3 at an optimal concentration (1 mg/L) promotes stomatal development and supports plant physiological activity, whereas higher concentrations tend to suppress stomatal formation as part of stress adaptation mechanisms.

The stomatal characteristics of *Dendrobium* sp. plantlets after GA_3 treatment are presented in Figure 2. The visualization clearly shows stomatal structures (A) surrounded by guard cells (B) and adjacent epidermal cells (C), allowing comparison of stomatal distribution among treatments.



Note: A = stomata; B = guard cells; C = epidermal cells.

Figure 2 Stomatal density on the leaf epidermis of *Dendrobium* sp. plantlets after GA_3 treatment (A-E)

Figure 2 illustrates differences in stomatal distribution on the leaf epidermis following GA_3 application. At higher GA_3 concentration (3 mg/L), stomata appeared less densely distributed, which is consistent with the quantitative data presented in Table 4. This observation suggests that increasing GA_3 concentration may reduce stomatal density and alter stomatal distribution patterns.

Such changes may influence transpiration regulation and water-use efficiency under drought stress conditions [31,32]. These findings are in line with previous reports indicating that GA_3 plays a role in stomatal development through hormonal crosstalk, particularly involving the GA–DELLA signaling pathway and its interaction with abscisic acid (ABA), which regulates stomatal formation and adaptive responses to environmental stress [28,33].

These findings provide a practical basis for optimizing *In vitro* drought stress simulation in orchid propagation systems.

4. Conclusion

The application of GA_3 at 1 mg/L was identified as the optimal concentration for supporting the growth of *Dendrobium* sp. plantlets under *In vitro* drought stress conditions. This was reflected by improved plantlet survival, better morphological performance, higher stomatal density, and increased chlorophyll content compared to higher GA_3 concentrations.

In contrast, excessive GA_3 application (≥2 mg/L) tended to reduce stomatal density and chlorophyll content, indicating a potential inhibitory effect under stress conditions. These findings highlight the dose-dependent role of GA_3, where optimal concentrations promote physiological adaptation, while higher concentrations may disrupt plant metabolic balance.

Therefore, the appropriate selection of GA_3 concentration is crucial to enhance plantlet tolerance to drought stress and to optimize *In vitro* culture performance. These findings provide a scientific basis for optimizing plant growth regulator application in orchid tissue culture under water-limited conditions.

Compliance with ethical standards

Acknowledgments

The authors would like to express their sincere gratitude to the Botanical Laboratory, particularly the *In vitro* Culture Room, Department of Biology, Faculty of Mathematics and Natural Sciences, University of Lampung, Bandar Lampung, Indonesia, for providing the facilities and technical support necessary to conduct this research.

Disclosure of conflict of interest

The authors declare that they have no conflicts of interest.

References

[1] Ahmad, N., Mehmood, M., Ali, S., & Abbas, Z. Effect of plant growth regulators on micropropagation of orchids: a review. International Journal of Agriculture and Biology. 2015; 17: 11–20.

[2] Kim, J.-S., & Kidokoro, S. Regulatory networks in plant responses to drought and cold stress. Plant Physiology. 2024; 195(1): 170–189.

[3] Huang, S., & Jin, S. Enhancing drought tolerance in horticultural plants through plant hormones: a strategic coping mechanism. Frontiers in Plant Science. 2025; 15: 1502438.

[4] Litvin, F. F., Ivanov, V. B., & Frolov, A. V. Drought stress-induced changes in gibberellin metabolism and stomatal development in tomato. Journal of Plant Growth Regulation. 2016; 35(3): 708–718.

[5] Nurcahyani, E., Hadisutrisno, B., Sumardi, I., & Suharyanto. Identification of Vanilla (*Vanilla planifolia* Andrews) Plantlet Lines Resistant to *Fusarium oxysporum* f. sp. *vanillae* Infection through *In vitro* Selection Using Fusaric Acid. Proceedings of the National Seminar of PFI Joglosemar Branch. 2014; 1(1): 272–279.

[6] Miazek, K., & Ledakowicz, S. Chlorophyll extraction from leaves using different solvents. Acta Agrophysica. 2002; 60**:** 75–88.

[7] Nurunisa, I., Sri, S., & Karyati. Comparison of stomatal density and stomatal pore size in several mangrove species in the Tongke-Tongke Coastal Area, Sinjai. Journal of Forests and Communities*. 2018*; 10(1): 67–75.

[8] Baskin, C. C., & Baskin, J. M. Seeds: Ecology, biogeography, and evolution of dormancy and germination (2nd ed.). 2014. Academic Press.

[9] Saeidnia, S., Gohari, A. R., & Manayi, A. Toxic effects of plant growth regulators. Pharmacognosy Reviews. 2017; 11(22): 145–150.

[10] Flematti, G. R., Waters, M. T., & Smith, S. M. Stereospecificity in gibberellin perception and response. Journal of Biological Chemistry. 2013; 288(28): 20017–20025.

[11] Khan, M. N., Zhang, J., Luo, T., & Xie, W. Regulation of plant growth hormones under abiotic stress conditions. Plant Physiology and Biochemistry. 2020; 151: 53–68.

[12] Chen, H., Li, Y., Yin, Y., Li, J., Li, L., Wu, K., Fang, L., & Zeng, S. Gibberlic Acid Inhibits Dendrobium nobile–Piriformospora Symbiosis by Regulating the Expression of Cell Wall Metabolism Genes. Biomolecules. 2023; 13(11): 1649.

[13] Kumari, S., & Singh, V. Role of gibberellins in plant growth regulation and stress responses: A review. Physiology and Molecular Biology of Plants. 2021; 27: 129–140.

[14] Podwyszyńska, M., Marasek-Ciolakowska, A., & Dziurka, M. Hormonal control of chlorophyll degradation and leaf senescence in micropropagated plants. Plant Growth Regulation. 2018; 84: 477–489.

[15] Gonçalves, S., Martins, J., & Romano, A. The role of plant growth regulators in micropropagation of ornamental plants: A review. Floriculture and Ornamental Biotechnology. 2019; 13: 1–12.

[16] Afzal, I., Javed, T., Farooq, M. (2022). Gibberellic acid-mediated hormonal regulation improves plant growth under stress conditions: A review. Plant Growth Regulation, 97, 197–212.

[17] Mohamed, M. A. H., & Alsadon, A. A. (2010). *In vitro* propagation of Solanum tuberosum L. and the effect of gibberellic acid on chlorosis and shoot quality. Journal of Plant Biochemistry and Biotechnology, 19(1), 1–7.

[18] Zhang, Y., Liu, X., Wang, M., & Li, H. (2023). Gibberellic acid modulates antioxidant defense systems and chloroplast integrity under abiotic stress in maize (Zea mays L.). Plant Physiology and Biochemistry, 194, 53–63.

[19] Kulus, D., & Zalewska, M. (2021). Phytotoxicity and other adverse effects in *In vitro* plant tissue cultures. Plants, 10(4), 670.

[20] Taiz, L., & Zeiger, E. (2015). Plant physiology and development (6th ed.). Sinauer Associates.

[21] Salisbury, F. B., & Ross, C. W. (1992). Plant physiology (4th ed.). Wadsworth Publishing.

[22] Hopkins, W. G., & Hüner, N. P. A. (2009). Introduction to plant physiology (4th ed.). John Wiley & Sons.

[23] Setiawan, A. N., Vistiadi, K., & Sarjiyah, S. (2021). Perkecambahan dan pertumbuhan bawang merah dengan direndam dalam giberelin. Jurnal Penelitian Pertanian Terapan, 21(1), 40–50

[24] Lestari, E. G. (2011). Peranan zat pengatur tumbuh dalam perbanyakan tanaman melalui kultur jaringan. Jurnal AgroBiogen, 7(1), 63–68.

[25] Phunthong, C., Pitaloka, M. K., Chutteang, C., Ruengphayak, S., Arikit, S., & Vanavichit, A. (2024). Rice mutants, selected under severe drought stress, show reduced stomatal density and improved water use efficiency under restricted water conditions. Frontiers in Plant Science, 15, 1307653.

[26] Nerva, L., Chitarra, W., & Gaiotti, F. (2023). Variability in stomatal adaptation to drought among grapevine cultivars: Genotype-dependent responses. Agriculture, 13(12), 2186.

[27] Caine, R. S., Harrison, E. L., Sloan, J., Flis, P. M., Fischer, S., Khan, M. S., Nguyen, P. T., Nguyen, L. T., Gray, J. E., & Croft, H. (2023). The influences of stomatal size and density on rice abiotic stress resilience. New Phytologist, 237(6), 2180–2195.

[28] Achard, P., Renou, J.-P., Berthomé, R., Harberd, N. P., & Genschik, P. (2008). Plant DELLAs restrain growth and promote survival of adversity by reducing the levels of reactive oxygen species. Current Biology, 18(9), 656–660.

[29] Liu, X., Yang, K., Qian, Y., Zhang, G., Yu, M., & Xie, Q. (2021). Gibberellin promotes stomatal development by mediating DELLA–SPCH interaction in Arabidopsis. Plant Physiology, 185(4), 1951–1965.

[30] Wei, H. (2021). Phytohormones and their crosstalk in regulating stomatal development and patterning. Journal of Experimental Botany, 72(7), 2356–2368.

[31] Wang, X., & Bergmann, D. C. (2018). Hormonal and environmental signals guiding stomatal development. BMC Biology, 16(1), 21.

[32] Zeng, L., Gowri, G., & Oosterhuis, D. M. (2022). Gibberellic acid improves photosynthetic electron transport and stomatal function in crops that are adversely affected by salinity exposure. Plants, 14(21), 3388.

[33] Kawaguchi, N., Iwase, A., & Ueguchi-Tanaka, M. (2014). GID1 modulates stomatal response and submergence tolerance involving abscisic acid and gibberellic acid signaling in rice. Journal of Experimental Botany, 65(6), 1635-1647.