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Optimizing Ex Vitro Acclimatization of Vanilla planifolia: Dynamics Effects of Substrate Composition and Indole-3-Butyric Acid (IBA)

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Abstract

The *ex-vitro* acclimatization of tissue-cultured *Vanilla planifolia* represents a critical propagation bottleneck, characterized by high mortality due to transplant shock and underdeveloped root systems. This study evaluated the interactive effects of organic substrate composition and exogenous Indole-3-butyric acid (IBA) on the morpho-physiological establishment of vanilla plantlets. A 4 × 4 factorial experiment was conducted using a Completely Randomized Design (CRD), testing four acclimatization media (raw rice husk, rice husk charcoal, cocopeat, and fern roots) alongside four IBA concentrations (0, 100, 200, and 300 ppm). Morphological data collected at eight weeks after transplanting revealed a distinct survival-growth paradox. Highly aerated substrates, such as raw rice husk, significantly promoted vertical shoot elongation (12.23 cm) but resulted in unsustainably low survival rates (33.33%) due to insufficient hydraulic buffering. Conversely, highly retentive media like cocopeat yielded a superior survival rate (63.89%). Crucially, the application of 200 ppm IBA emerged as the definitive biochemical optimum. This targeted concentration successfully broke morphological dormancy to maximize root organogenesis, producing an average of 5.00 roots at 6.09 cm in total length. This robust root architecture functioned as a physiological sink, driving maximum leaf expansion and total fresh biomass accumulation (2.28g), whereas higher concentrations (300 ppm) induced phytotoxicity. Ultimately, this study demonstrates that combining a highly moisture-retentive substrate with a 200 ppm IBA application effectively resolves the survival-growth trade-off. This research delivers a highly reproducible, mechanistic protocol to minimize acclimatization mortality and significantly accelerate the scalable production of vigorous vanilla planting materials for the global agricultural industry.

1. Introduction

Vanilla planifolia is a highly valued horticultural crop cultivated primarily for its cured beans, which yield vanillin, a crucial flavorings and aromatic compound in the global food, beverage, cosmetic, and pharmaceutical industries [1]. Driven by high global demand, vanilla prices range from 80 to 175 USD per kilogram. Indonesia currently stands as the world's second-largest vanilla producer after Madagascar [2]. To capitalize on this economic potential and elevate its export capacity, there is a pressing need to optimize mass propagation techniques to supply high-quality, disease-free planting materials.

In vitro micropropagation offers a viable solution for the rapid multiplication of vanilla seedlings, the subsequent ex vitro acclimatization phase remains a significant propagation bottleneck. During this stage, plantlets must undergo a critical physiological transition from a heterotrophic or mixotrophic in vitro environment, characterized by high humidity, low light, and external carbon sources, to an entirely autotrophic existence in the greenhouse [3]. This abrupt transition to *ex vitro* conditions often results in high mortality rates, primarily driven by severe transplant shock and rapid desiccation. Because plantlets develop in a high-humidity, heterotrophc *in vitro* environment, they exhibit altered anatomy. Their leaves possess underdeveloped epicuticular waxes and structurally abnormal, sluggish stomata that fail to close efficiently in response to lowered ambient humidity, leading to unchecked transpiration water

loss. Concurrently, it's *in vitro*-formed root systems are fragile, typically lack root hairs, and have poor vascular connections to the shoot. Consequently, these roots are highly inefficient at water and nutrient uptake, creating a lethal imbalance where rapid foliar water loss vastly outpaces the roots' hydraulic capacity. Consequently, facilitating rapid root induction and mitigating transplant shock through optimized substrate selection and exogenous hormonal application is imperative for plantlet survival.

The physical and chemical properties of the acclimatization substrate play a vital role in root establishment. Organic media such as rice husk charcoal, cocopeat, and fern roots are frequently utilized due to their excellent porosity, aeration, and moisture retention capabilities [4]. Previous studies on epiphytic and semi-epiphytic species, including Dendrobium and Vanda orchids, have demonstrated that porous media like cocopeat and fern roots significantly enhance survival rates, biomass accumulation, and root volume [5], [6]. Specific to vanilla, fern roots (Kadaka) have been reported to support up to a 90% survival rate during early acclimatization while promoting rapid leaf area expansion [7], [8]. However, relying solely on physical substrates is often insufficient to overcome the initial rooting delay characteristic of tissue-cultured vanilla.

Stimulation of root organogenesis, induced by the application of exogenous auxins, particularly Indole-3-butyric acid (IBA), is widely adopted. IBA is highly effective in promoting adventitious root formation, accelerating root elongation, and enhancing nutrient absorption capacity during the critical acclimatization window. Research on related horticultural species has established that IBA application—typically ranging from 100 to 200 mg/L— The significant improvement in plantlet vigour is directly driven by the biochemical role of IBA in regulating meristematic activity. IBA functions as a stable auxin precursor, providing a slow-release pool of active IAA that avoids the rapid enzymatic degradation typical of endogenous auxins. This sustained auxin signalling activates the pericycle, stimulating intensive cellular dedifferentiation and elongation to form adventitious roots. Consequently, the enhanced root architecture maximizes hydraulic conductivity and nutrient assimilation. This rapid resource acquisition relieves *ex vitro* transplantation stress, upregulates photosynthetic capacity, and provides the necessary resources to fuel increased leaf expansion and superior overall plantlet vigour [3], [9]. Despite these proven benefits, the specific synergistic effects of varying organic substrates combined with IBA concentrations on V. planifolia acclimatization remain underexplored.

Given the critical role of both the physical growth medium and hormonal regulation, this study aims to evaluate the interactive effects of different organic acclimatization media and varying concentrations of exogenous IBA on the morpho-physiological growth and survival of tissue-cultured vanilla plantlets. By identifying the optimal substrate-hormone combination, this research seeks to establish a standardized, high-efficiency protocol for vanilla seedling production, ultimately contributing to the sustainable expansion of the vanilla industry in Indonesia.

2. Research Methods

2.1. Study Site and Plant Material

The acclimatization experiment was conducted from July to September 2024 within a climate-controlled greenhouse at the Plantation Seed Centre (BBTP Salatiga, Central Java, Indonesia). The facility is located at an elevation of 606.96 m above sea level, maintaining a mean daily temperature of 29.67 °C and an average relative humidity of 57%. Healthy, uniform in vitro vanilla (*Vanilla planifolia*) plantlets, characterized by the presence of 3 to 4 roots and 4 to 5 fully expanded leaves, were selected as the starting material for this study.

2.2. Preparation of Acclimatization Substrates and Hormonal Treatments

The basal acclimatization substrate consisted of a homogenous mixture of red soil, black soil, and composted goat manure. Four distinct media combinations were prepared by adding different organic amendments to the basal mixture in a 1:1:1:1 (v/v/v/v) ratio: M0: Raw rice husk + basal mixture; M1: Rice husk charcoal + basal mixture; M2: Cocopeat + basal mixture; M3: Fern roots (Kadaka) + basal

mixture. All prepared substrates were placed into perforated 16-oz plastic containers and pre-treated with a 2 g/L systemic fungicide solution to prevent soil-borne pathogens. Indole-3-butyric acid (IBA) stock solutions were prepared by dissolving the exogenous auxin powder in a minimal volume of NaOH (Author Note: specify concentration here, e.g., 1 N NaOH) before diluting to a 1 L final volume with distilled water. Serial dilutions were subsequently performed to achieve the four targeted treatment concentrations: 0 (control), 100, 200, and 300 mg/L.

2.3. Acclimatization Procedure

Prior to transplanting, plantlets were carefully extracted from their culture vessels, and the root systems were gently washed under running water to remove residual agar. To mitigate fungal infection risks, the plantlets were submerged in a 2 g/L fungicide solution for 10 minutes. Subsequently, the basal portions of the plantlets were dipped into their respective assigned IBA treatment solutions for 10 minutes. The treated plantlets were transplanted into the prepared substrate containers and immediately covered with transparent plastic hoods to maintain a high-humidity microclimate. Moisture levels were maintained via sub-irrigation (bottom watering) three times per week. After a two-week transitional period, the plastic hoods were gradually removed to expose the plantlets to ambient greenhouse conditions. Routine maintenance included daily microclimate monitoring, manual weed eradication, and the prompt removal of any necrotic plantlets.

2.4. Experimental Design and Data Collection

The study was arranged in a Completely Randomized Design (CRD) using a 4 × 4 factorial scheme. The first factor was the growth medium composition (M0, M1, M2, M3), and the second factor was the IBA concentration (0, 100, 200, and 300 mg/L). The experiment comprised 16 treatment combinations, each replicated three times. Every experimental unit contained three plantlets, yielding a total of 144 observed plants. Growth and morphological parameters were systematically recorded at the conclusion of the observation period (Author Note: specify exact timing here, e.g., 8 weeks after transplanting (WAT)). The measured variables included plant survival rate (%), leaf number, leaf length (cm), leaf width (cm), primary root count, root length (cm), and total plantlet fresh biomass (g).

2.5. Statistical Analysis

Data were subjected to an Analysis of Variance (ANOVA) at a 5% significance level ($\alpha = 0.05$). Where treatment effects or interactions were found to be statistically significant, means were separated using Fisher's Least Significant Difference (LSD) post-hoc test at $P \leq 0.05$.

3. Results and Discussions

The ex-vitro acclimatization of tissue-cultured Vanilla planifolia represents a critical physiological bottleneck, characterized by high mortality due to the abrupt shift from a highly controlled, high-humidity in vitro environment to a fluctuating autotrophic state. As demonstrated in Table 1, early establishment and plantlet survival are fundamentally governed by the physical architecture and matric potential of the acclimatization substrate. Our data reveal that raw rice husk (M0) yielded an unsustainably low mean survival rate (33.33%). While highly aerated, macro-porous substrates allow for rapid water drainage and root respiration, their poor moisture retention makes them unsuitable as a standalone medium during the early stages of acclimatization. Macro-porous substrates prevent hypoxia, their low water-holding capacity fails to buffer against the severe vapor pressure deficits encountered during greenhouse transfer [10].

Conversely, cocopeat (M2) demonstrated a statistically superior mean survival rate (63.89%), culminating in 100% survival when integrated with higher concentrations of Indole-3-Butyric Acid (IBA). This aligns with Ayuningtyas et al. [11] who established that substrate moisture retention is the most critical factor preventing lethal desiccation before stomatal competence is fully developed. The fibrous, highly porous organic structure of coconut coir exhibits exceptional matric potential, effectively preserving moisture while retaining exogenously applied hormones in the root zone [12]. Consequently,

Table 1 establishes the baseline prerequisite for vanilla acclimatization: hydraulic buffering via substrate selection is the primary safeguard against transplant shock. Because in vitro plantlets lack functional stomatal regulation, the physical matrix of the substrate must act as an external buffer. Media that maintain consistent capillary moisture compensate for the plantlet's inefficient water uptake, preventing the rapid desiccation observed in highly aerated but dry substrates.

Substrate physics governed hydration and baseline survival, while biochemical intervention was absolutely essential to break the morphological dormancy of the root system. Quantitative metrics in Table 2, corroborated by visual phenotyping (Figures 1 and 2), unequivocally demonstrate that untreated controls (0 ppm IBA) exhibited rudimentary, developmentally arrested roots across all media types, yielding the lowest root counts (2.86 ± 0.64) and shortest lengths (3.73 ± 0.48 cm). The application of 200 ppm exogenous IBA induced a profound morphogenetic response, representing a distinct biochemical optimum. This treatment yielded statistically maximum root counts (5.00 ± 0.41) and root lengths (6.09 ± 0.34 cm). This extensive proliferation is distinctly visible in the morphological tracking (Figure 1), where the 200 ppm cohorts exhibit dense, highly elongated root networks. The qualitative contrast is further emphasized in Figure 2, highlighting the striking emergence of both basal and adventitious root primordia compared to the sparse root architecture of the control group.

Table 1. Survival rates of vanilla plantlets in response to varying growth media and IBA concentration at 8 WAT.

Growth Media	IBA Concentration (ppm)				Means
	Control	100	200	300	
	---------- (%) ----------				
Rice Husk	44.44	33.33	33.33	22.22	33.33 ± 9.071^{b}
Rice Husk Charcoal	44.44	55.56	22.22	55.56	44.44 ± 15.717^{ab}
Cocopeat	44.44	44.44	66.67	100.00	63.89 ± 26.257^{a}
Fern Root	44.44	66.67	44.44	33.33	47.22 ± 13.984^{ab}
Rata-Rata	44.44	50.00	41.66	52.77	

Means within a column followed by different superscripts are significantly different (P < 0.05).

Table 2. Effects of various acclimatization media and Indole-3-butyric acid (IBA) concentrations on the root development of vanilla plantlets at 8 WAT.

Treatments	Number of Roots	Root Length (cm)
Growth Media		
Rice Husk	4.02 ± 1.23	4.91 ± 1.01
Rice Husk Charcoal	3.95 ± 0.73	4.67 ± 0.75
Cocopeat	4.11 ± 0.91	4.79 ± 1.29
Fern Root	3.67 ± 1.16	4.72 ± 1.43
IBA Concentration		
0 ppm	2.86 ± 0.64^{c}	3.73 ± 0.48^{c}
100 ppm	3.96 ± 0.34^{ab}	4.98 ± 0.70^{ab}
200 ppm	5.00 ± 0.41^{a}	6.09 ± 0.34^{a}
300 ppm	3.92 ± 0.80^{bc}	4.30 ± 0.60^{bc}

Means within a column followed by different superscripts are significantly different (P < 0.05).

The mode of action underpinning this rapid organogenesis relies on auxin's ability to trigger cellular dedifferentiation in the pericycle. Exogenous IBA is basipetally transported and slowly metabolized into Indole-3-acetic acid (IAA). Following the acid-growth theory [13], this auxin gradient stimulates plasma membrane H+-ATPases, acidifying the apoplast and activating expansions that cleave calcium-pectin

cross-links in the cell wall. This loosening drives the rapid cell expansion and active meristematic division visually captured in Figures 1 and 2 [14], [15]. Crucially, Table 2 also highlights a biphasic, dose-dependent regulatory threshold. At 300 ppm IBA, root metrics regressed significantly. This supra-optimal phytotoxicity is governed by auxin-ethylene crosstalk: excessive intracellular auxin upregulates 1-aminocyclopropane-1-carboxylate (ACC) synthase, triggering a localized ethylene burst that aggressively inhibits root meristematic activity.

The successful establishment of a robust root architecture (detailed in Table 2 and visually confirmed in Figures 1 and 2) had profound downstream effects on canopy development, validating a critical sink-source physiological model. By maximizing root proliferation via the 200 ppm IBA treatment, the plantlets drastically expanded their subterranean foraging network. This enhanced root system acted as a highly efficient "sink" for water and mineral acquisition [16].

Once absorbed, these resources were rapidly translocated via the xylem to the developing aerial tissues, satisfying the high transpiration demand and supporting new vegetative growth. The continuous influx of water and dissolved nutrients sustained stomatal conductance and high photosynthetic rates. As noted by Lambers on [17], lateral and longitudinal foliar growth is heavily dictated by the availability of photosynthetic assimilates, which provide the carbon skeletons and energy requisite for cellular division. Reflecting this physiological cascade, Table 3 demonstrates that plantlets treated with 200 ppm IBA exhibited the most significant foliar expansion at 8 weeks after transplanting (WAT), achieving statistical maximums in both leaf width (1.43 ± 0.24 cm) and leaf length (4.05 ± 0.47 cm).

The synthesis of our physical and biochemical interventions culminates in the final architectural metrics presented in Table 4, plant height and total fresh weight. Here, a fascinating physiological dichotomy emerges between substrate aeration and hormonal optimization. Despite raw rice husk (M0) yielding the lowest survival rates (Table 1), it significantly promoted vertical shoot elongation, yielding the tallest surviving plantlets (12.23 ± 1.84 cm; Table 4). The mechanistic advantage here lies in its high porosity, which drastically reduces bulk density and improves rhizosphere aeration. This high oxygen diffusion rate sustains mitochondrial respiration, generating the ATP requisite for active nutrient transport and rapid vertical stem elongation. However, total biomass accumulation—represented by fresh weight—tells a different, more holistic story. The 200 ppm IBA treatment, which optimized root organogenesis (Table 2, Figures 1 & 2) and canopy expansion (Table 3), ultimately drove the highest total fresh biomass accumulation (2.28 ± 0.15 g; Table 4). This corroborates Nikmah et al. [18], who linked fresh weight directly to total organs expansion and optimized internal hydration capacities.

Table 3. Effects of various growth media and Indole-3-butyric acid (IBA) concentrations on the leaf morphology of vanilla plantlets during acclimatization.

Treatments	Number of Leaves		Leaf Width (cm)		Leaf Length (cm)	
	2 WAT	8 WAT	2 WAT	8 WAT	2 WAT	8 WAT
Growth Media						
Rice husk	3.53 ± 0.28	3.32 ± 0.66	0.91 ± 0.19	1.02 ± 0.22	3.06 ± 0.36	3.5 ± 0.73
Rice husk charcoal	3.88 ± 0.55	3.75 ± 0.32	0.98 ± 0.11	1.15 ± 0.28	2.9 ± 0.31	3.49 ± 0.85
Cocopeat	3.61 ± 0.46	4.01 ± 0.09	0.99 ± 0.12	1.08 ± 0.12	3.03 ± 0.21	3.46 ± 0.22
Fern roots	3.47 ± 0.28	3.58 ± 0.59	0.94 ± 0.11	1.15 ± 0.37	2.93 ± 0.39	3.3 ± 0.40
IBA Concentration						
0 ppm	3.72 ± 0.26^{a}	3.51 ± 0.59	0.93 ± 0.15^{ab}	0.94 ± 0.07^{b}	2.91 ± 0.32^{ab}	3.01 ± 0.31^{b}
100 ppm	3.14 ± 0.11^{b}	3.47 ± 0.68	0.83 ± 0.06^{b}	0.96 ± 0.09^{b}	2.72 ± 0.11^{b}	3.06 ± 0.33^{b}
200 ppm	3.87 ± 0.46^{a}	3.95 ± 0.19	1.06 ± 0.12^{a}	1.43 ± 0.24^{a}	3.28 ± 0.18^{a}	4.05 ± 0.47^{a}
300 ppm	3.75 ± 0.28a	3.74 ± 0.42	1.00 ± 0.09^{a}	1.08 ± 0.13^{b}	3.02 ± 0.29^{a}	3.64 ± 0.29^{a}

Means within a column followed by different superscripts are significantly different ($P < 0.05$).

Fig 1. Visual assessment of root morphology across different substrate compositions and exogenous IBA treatments at 8 weeks post-acclimatization. *(Red markers indicate specific root development)*

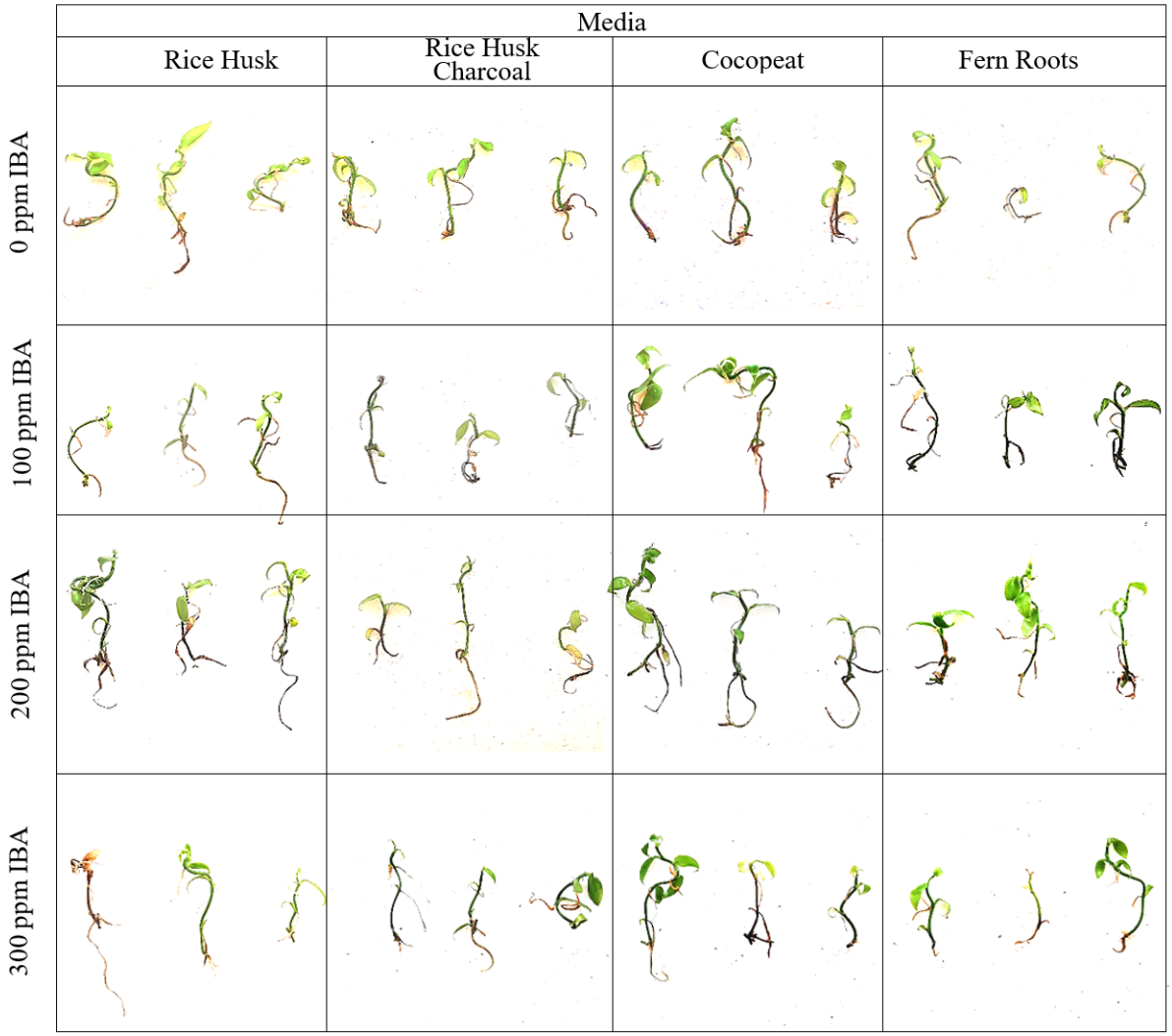

Fig 2. Representative phenotypic responses of acclimatized vanilla plantlets subjected to different substrate compositions and exogenous IBA treatments.

Table 4. Growth of acclimatized vanilla plants with the application of different growth media acclimatization and IBA concentrations at 8 WAT

Treatments	Plant height (cm)	Fresh weight (g)
Growth Media		
Rice husk	12.23 ± 1.84^{a}	1.77 ± 0.38
Rice husk charcoal	10.15 ± 1.42^{b}	1.92 ± 0.40
Cocopeat	8.98 ± 1.19^{b}	2.01 ± 0.81
Fern roots	9.44 ± 1.32^{b}	1.49 ± 0.71
IBA Concentration		
0 ppm	8.88 ± 1.54^{c}	1.28 ± 0.39^{c}
100 ppm	9.89 ± 0.89^{ab}	2.06 ± 0.73^{ab}
200 ppm	11.39 ± 0.89^{a}	2.28 ± 0.15^{a}
300 ppm	10.64 ± 2.88^{ab}	1.56 ± 0.36^{bc}

Means within a column followed by different superscripts are significantly different ($P < 0.05$).

The novelty of this study lies in the explicit uncoupling—and subsequent sequential integration—of the physical and biochemical drivers of vanilla acclimatization. We demonstrate that relying solely on a highly aerated substrate (like rice husk) prioritizes vertical growth but incurs devastating mortality, whereas highly retentive substrates (like cocopeat) ensure survival but require chemical calibration to optimize growth. By establishing that combining a highly retentive matrix with a brief, targeted exposure to 200 ppm IBA forces rapid root organogenesis, subsequently driving canopy expansion and maximum biomass accumulation, this study provides a highly reproducible protocol. Implementing this specific combinatorial matrix will significantly truncate the nursery acclimatization phase, lower mortality-associated economic losses, and ensure the scalable deployment of vigorous *Vanilla planifolia* clones to commercial plantations.

4. Conclusion

The successful ex vitro establishment of Vanilla planifolia fundamentally relies on a dynamic physiological synergy between the structural hydrology of the acclimatization matrix and the precise calibration of exogenous phytohormones. Our findings expose a critical survival-growth paradox: highly aerated substrates, such as raw rice husk (M0), drive significant vertical shoot elongation by sustaining root respiration but fail to provide the hydraulic buffering necessary to avert high mortality. Conversely, highly retentive media, like cocopeat and rice husk charcoal, successfully safeguard plantlet viability against transplant shock but lack the physical stimuli for rapid elongation. To fully realize the agronomic potential of this optimized acclimatization protocol, future research must advance beyond controlled greenhouse conditions to encompass both molecular and longitudinal field evaluations. Initially, transcriptomic analyses are necessary to elucidate the precise molecular mechanisms by which the 200 ppm IBA optimum induces rapid root organogenesis, and to identify the specific pathways mediating phytotoxicity at elevated concentrations. Finally, rigorous field trials are essential to validate this methodology. It remains to be determined whether the rapid root establishment and early biomass accumulation stimulated by the 200 ppm IBA treatment will translate into sustained vine vigour, advanced flowering, and enhanced pod yield under commercial plantation conditions.

References:

[1] H. S. Abdat, S. I. Santoso, and S. Nurfadillah, "Daya saing komoditas vanili Indonesia di pasar internasional," *Jurnal Ekonomi Pertanian dan Agribisnis*, vol. 6, no. 3, pp. 1084–1097, 2022. https://doi.org/10.21776/ub.jepa.2022.006.03.28

[2] Food and Agriculture Organization of the United Nations, "Top 10 country, export quantity of vanilla," 2022. [Online]. Available: http://www.fao.org/faostat/en/#rankings/countriesbycommodityexports. [Accessed: May 15, 2024].

[3] A. Astutik, A. Sumiati, and S. Sutoyo, "Stimulasi pertumbuhan *Dendrobium* sp menggunakan hormon auksin naphtalena acetic acid (NAA) dan indole butyric acid (IBA)," *Buana Sains*, vol. 21, no. 1, pp. 19–28, 2021. https://doi.org/10.33366/bs.v21i1.2659

[4] V. Andriani and I. A. K. Pramushinta, "Pengaruh media tanam terhadap respon fisiologi aklimatisasi anggrek Cattleya," *STIGMA: Jurnal Matematika dan Ilmu Pengetahuan Alam UNIPA*, vol. 10, no. 1, pp. 17–19, 2017. https://doi.org/10.36456/stigma.vol10.no1.a1469

[5] T. Sudartini, D. Zulmani, and D. Diantini, "Pengaruh sungkup dan jenis media tanam terhadap pertumbuhan bibit anggrek Dendrobium saat aklimatisasi," *Media Pertanian*, vol. 5, no. 1, pp. 31–43, 2020.

[6] A. F. Nuzullah and R. Firgiyanto, "Aplikasi berbagai jenis media dan zpt terhadap aklimatisasi anggrek vanda (*Vanda* sp.)," in *National Conference Proceedings of Agriculture*, Politeknik Negeri Jember, 2021, pp. 10–24. https://doi.org/10.25047/agropross.2021.202

[7] E. W. Tini, P. Sulistyanto, and G. H. Sumartono, "Aklimatisasi anggrek (*Phalaenopsis amabilis)* dengan media tanam yang berbeda dan pemberian pupuk daun," *Jurnal Hortikultura Indonesia*, vol. 10, no. 2, pp. 119–127, 2019. http://dx.doi.org/10.29244/jhi.10.2.119-127

[8] B. M. Dewi, D. Nurhaliza, N. Aprilia, P. Handayani, and W. Sari, "Pengaruh media tanam terhadap aklimatisasi planlet anggrek *Dendrobium* sp di UPTD Balai Perbanyakan Benih Tanaman Pangan Hortikultura Provinsi Sumatera Selatan. In *Prosiding Seminar Nasional Biologi* (Vol. 1, No. 1, pp. 539-548).

[9] W. Fikrinda and A. Astutik, "Pengaruh jenis dan konsentrasi auksin terhadap pertumbuhan anggrek *Dendrobium* sp pada fase aklimatisasi," Bachelor Thesis, Univ. Tribhuwana Tunggadewi, 2022.

[10] A. A. Fauziah, N. Prahasti, N. Setiari, and E. Saptiningsih, "Naungan dan tipe substrat berbeda pada periode aklimatisasi ex-vitro *Phalaenopsis* hibrid," *Buletin Anatomi dan Fisiologi*, vol. 5, no. 1, pp. 60–66, 2020.

[11] U. Ayuningtyas, B. Budiman, and T. K. K. Azmi, "Pengaruh pupuk daun terhadap pertumbuhan bibit anggrek Dendrobium Dian Agrihorti pada tahap aklimatisasi," *Jurnal Pertanian Presisi*, vol. 4, no. 2, pp. 148–159, 2021. https://doi.org/10.35760/jpp.2020.v4i2.2888

[12] T. Rahayu, G. E. Jayanti, and D. Agisimanto, "Indole-3-butyric acid induced adventitious root of *Dendrobium Milla Nayla x Dendrobium striaenopsis* planted on coco-husk and wood charcoal during acclimatization stage," *Berkala Penelitian Hayati*, vol. 28, no. 1, pp. 39–43, 2022. https://doi.org/10.23869/bphjbr.28.1.20226

[13] R. Asra, R. A. Samarlina, and M. Silalahi, *Hormon Tumbuhan*. Jakarta: UKI Press, 2020.

[14] D. Widiastoety, "Pengaruh auksin dan sitokinin terhadap pertumbuhan planlet anggrek Mokara (Effect of auxin and cytokinin on the growth of Mokara orchid plantlets)," *Jurnal Hortikultura*, vol. 24, no. 3, pp. 230–238, 2014.

[15] A. W. Wheeler and E. C. Humphries, "Effect of gibberellic acid on growth, gibberellin content, and chlorophyll content of leaves of potato (*Solanum tuberosum*)," *Journal of Experimental Botany*, vol. 14, no. 1, pp. 132–136, 1963. https://doi.org/10.1093/jxb/14.1.132

[16] H. R. Fitri, T. Rahayu, G. E. Jayanti, and D. Agisimanto, "Pengaruh variasi konsentrasi indole butyric acid (IBA) terhadap pertumbuhan anggrek (*Dendrobium canaliculatum*) pada media tumbuh moss putih" *Jurnal Ilmiah Biosaintropis*, vol. 9, no. 2, pp. 38–46, 2024. https://doi.org/10.33474/e-jbst.v9i2.507

[17] H. Lambers and R. S. Oliveira, "Growth and Allocation," in *Plant Physiological Ecology*. Cham: Springer, 2019, ch. 10. https://doi.org/10.1007/978-3-030-29639-1_10

[18] Z. C. Nikmah, W. Slamet, and B. A. Kristanto, "Aplikasi silika dan NAA terhadap pertumbuhan anggrek bulan (*Phalaenopsis amabilis* L.) pada tahap aklimatisasi," *Jurnal Agro Complex*, vol. 1, no. 3, pp. 101–110, 2017. https://doi.org/10.14710/joac.1.3.101-110