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Effects of Trichoderma Doses and Coconut Water as Natural Plant Growth Regulator on the Vegetative Growth of Lime Cuttings

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ABSTRACT

Vegetative propagation through cuttings is relatively simple; however, lime cuttings (*Citrus aurantiifolia*) often lack roots capable of supporting upright growth. Natural plant growth regulators such as coconut water (auxins/cytokinins) stimulate rooting, while Trichoderma sp. enhances root development, nutrient absorption, and resistance to pathogens. This study evaluates the effect of Trichoderma dosage and coconut water concentration on the growth of lime cuttings. The experiment used a 3 × 4 factorial randomized block design: Trichoderma (0, 25, 50 g plant^{-1}) and coconut water (100%, 15%, 25%, 35% v/v) in a Green House (April–July 2025). Trichoderma and coconut water significantly affected the time to bud emergence and number of leaves, but their interaction was not significant for these traits; neither had an effect on the number of shoots. The fastest bud emergence occurred with 50 g plant^{-1} Trichoderma (7.9 days) and 100% coconut water (7.89 days), while 15% coconut water delayed it (10.67 days). Trichoderma 50 g plant^{-1} increased shoot length (10.9 cm) and leaf number (9.1) compared to the control (9.0 cm; 6.9 leaves). Coconut water 100% maximized shoot length (11.0 cm) and leaves (9.3) compared to 15% (9.0 cm; 6.7 leaves). Success reached 100% with combinations of Trichoderma 25 or 50 g and 100% coconut water. Application of Trichoderma 50 g plant^{-1} and undiluted coconut water effectively accelerated early shoot growth and improved the performance of lime cuttings.

INTRODUCTION

Plant propagation through cuttings is an efficient and straightforward method for seed propagation, with successful outcomes marked by the development of roots and shoots (Muleo et al., 2025). To expedite root and shoot growth in lime cuttings, the application of plant growth regulators (PGRs) is effective (Domingues Neto et al., 2024; Guchhait et al., 2024). These organic compounds can physiologically stimulate, inhibit, or modify plant growth and development both qualitatively and quantitatively. PGRs may be derived naturally or synthesized; for instance, natural PGRs can be extracted from coconut water (Kumar et al., 2024).

In contrast, pepper cultivation typically involves climbing pepper plants, which necessitate climbing poles, thereby increasing production costs. Additionally, the availability of climbing poles is becoming increasingly limited (Padilla et al., 2024). An alternative approach is the cultivation of shrub pepper plants, which do not require climbing poles due to the absence of clinging tendrils (Ahmad et al., 2024). Shrub pepper offers several benefits, including easier maintenance and harvesting, as well as earlier and continuous fruiting (Madhushani et al., 2024).

Coconut water, when used as a growth regulator, promotes root and shoot formation due to its content of auxin and cytokinin hormones. Thus, selecting the appropriate concentration is essential for the effective use of growth regulators. PGRs are efficacious at specific concentrations; excessive concentrations can harm the plant by causing excessive cell division and inhibiting root growth. Conversely, suboptimal hormone concentrations result in ineffective plant growth (Carganilla et al., 2025).

A challenge in propagating lime seedlings from stem cuttings is the insufficient root density to support upright growth (Swelih & Said, 2023). To produce robust lime seedlings, the biological control agent

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Trichoderma sp., a fungus from the Ascomycetes class with antifungal properties, can be utilized. Trichoderma sp. enhances root growth, protects against soil-borne and water-borne pathogens, improves plant vigor, and stimulates nutrient uptake when present in plant roots (Triasih & Widyaningsih, 2023). Indicates that Trichoderma also positively affects shoot length, shoot number, and accelerates seedling emergence.

Based on the above description, this study aims to evaluate the effect of Trichoderma dosage and coconut water concentration as natural growth regulators on the vegetative growth of lime cuttings (*Citrus aurantiifolia*), particularly in accelerating bud emergence, increasing the number of leaves, shoot length, and the success rate of lime cutting growth.

MATERIALS AND METHODS

This study was conducted from April to July 2025 at the Greenhouse of the Faculty of Agriculture, Muslim University of Indonesia. The materials used in this research included soil, rice husk charcoal, compost, Trichoderma, coconut water, clear plastic, label boards, lime cuttings, and polybags measuring 20 x 25 cm. The tools employed comprised pruning shears, shovels, hoes, rulers, books, pens, measuring cups, and scales. A randomized block design with a factorial pattern was implemented, consisting of two factors. The first factor involved the application of Trichoderma sp at three levels: no Trichoderma, 25 g per plant, and 50 g per plant. The second factor concerned the concentration of coconut water at four levels: 100% (1000 ml coconut water) as the control, 15% (150 ml coconut water) combined with 85% (850 ml plain water), 25% (250 ml coconut water) combined with 75% (750 ml plain water), and 35% (350 ml coconut water) combined with 65% (650 ml plain water). Each treatment was replicated three times, resulting in a total of 36 experimental units.

RESULTS AND DISCUSSION

1. Time for shoots to appear

Observational results regarding the emergence time of shoots from lime cuttings treated with Trichoderma and coconut water were analyzed. The analysis of variance indicated that both Trichoderma and coconut water individually had a highly significant impact. However, the interaction between Trichoderma and coconut water did not significantly influence the emergence time of shoots from lime cuttings.

Table 1. The mean duration for the emergence of shoots in lime seedlings following the application of Trichoderma and coconut water

Trichoderma sp.	Coconut water concentration				Average
	A0	A1	A2	A3	
T0	10.0	13.0	11.7	10.0	11.2[a]
T1	7.0	9.7	9.3	7.3	8.3[b]
T2	6.7	9.3	8.3	7.3	7.9[b]
Average	7.89[b]	10.67[a]	9.78[ab]	8.22[b]	
HSD (A)			2.0		
HSD (T)			1.6		

Note: Numbers followed by distinct letters within the same row (a, b, and c) and column (x, y, and z) exhibit significant differences at the 0.05% level according to the HSD test.

The 5% HSD test results presented in Table 1 reveal that applying Trichoderma at a rate of 50 g per plant (T2) significantly hastened shoot emergence, averaging 7.9 days after planting (dap). In contrast, the treatment without Trichoderma (T0) exhibited the slowest shoot emergence at 11.2 Day After Planting (DAP). This suggests that Trichoderma application may enhance early plant growth by improving nutrient availability or stimulating plant growth hormones (Gupta, 2020; Subramaniam et al., 2022). The soaking treatment with 100% coconut water (A0) led to the quickest shoot emergence at 7.89 dap, differing significantly from all other treatments except for soaking in 35% coconut water (A3). On the other hand, soaking with 15% coconut water (A1) resulted in the slowest shoot emergence at 10.67 dap. This indicates that a high concentration of coconut water positively influences the germination process and early shoot growth, likely due to the optimal natural hormone and nutrient content at full concentration

(Konappa et al., 2020; Korlina et al., 2023; Subramaniam et al., 2022).

Combining Trichoderma application with the right concentration of coconut water can optimize shoot emergence acceleration in plants. The T2 and A0 treatments demonstrate synergistic potential in promoting faster early growth compared to the control. These insights are valuable for cultivation practices focused on improving planting time efficiency and enhancing early plant (Karaca & Eltem, 2024; Srivastava et al., 2025).

2. Number of shoots

Observational results regarding the number of shoots from lime cuttings treated with Trichoderma and coconut water reveal that neither the application of Trichoderma nor coconut water, nor their interaction, significantly affected the shoot count. This conclusion is corroborated by the analysis of variance.

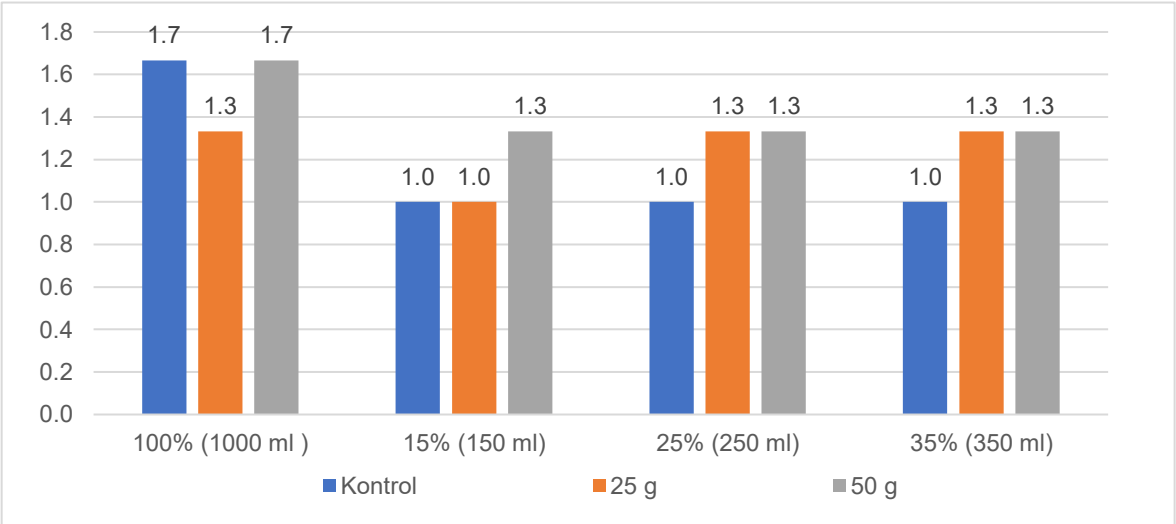

Figure 1. Average number of shoots of lime seedlings in the application of Trichoderma and coconut water at 12 Weeks After Planting (WAP)

Figure 1 shows that the treatment without trichoderma and 100% coconut water immersion (T0A0) and the treatment of 50g Trichoderma application and 100% coconut water immersion (T2A0) gave results that tended to be better on the number of shoots of lime seedling cuttings, namely 1.7 shoots. Treatments T0A1, T0A2, T0A3 showed the lowest number of shoots, namely 1.0 shoots.The results in Figure 1 indicate that the combination of no Trichoderma application with 100% coconut water immersion (T0A0) and 50g Trichoderma application with 100% coconut water immersion (T2A0) produced the highest average number of shoots per lime seedling cutting (1.7 shoots). This suggests that immersion in full-strength coconut water plays a significant role in promoting shoot proliferation, likely due to its rich nutrient and growth hormone content, which enhances cell division and elongation in the cuttings (Indriyanti et al., 2020; Shifka & Seran, 2025)

The fact that T2A0 (with Trichoderma) performed comparably to T0A0 (without Trichoderma) implies that while Trichoderma may contribute beneficial effects such as disease resistance or root growth promotion, its impact on shoot number under these conditions may be less pronounced than the effect of coconut water immersion (Sparta & Emilda, 2020). It is possible that the bioactive compounds in coconut water create an optimal physiological environment for shoot induction, overshadowing the additional benefit from Trichoderma at the tested concentration (Jayawardena et al., 2021; Nikila et al., 2025)

Conversely, treatments T0A1, T0A2, and T0A3, which presumably involved lower concentrations or absence of coconut water immersion combined with no Trichoderma application, resulted in the lowest shoot numbers (1.0 shoot) (Wilms et al., 2022). This highlights the critical role of coconut water concentration in stimulating shoot development. The reduced shoot number in these treatments could be attributed to insufficient availability of growth-promoting substances, leading to limited cell differentiation and shoot initiation (Kong et al., 2025; Wong et al., 2024). These findings emphasize the importance of

coconut water immersion in enhancing shoot proliferation in lime seedling cuttings. Trichoderma may offer benefits, but its effect on shoot number appears secondary to coconut water concentration. Further studies could examine interactive effects on shoot development.

3. Shoot length

The study investigated the shoot length of lime cuttings subjected to treatments with Trichoderma and coconut water. The analysis of variance indicated that both Trichoderma and coconut water individually had a significant impact on the shoot length of lime seedlings. However, the interaction between Trichoderma and coconut water did not exhibit a significant effect on the shoot length of the seedlings.

Table 2. Mean shoot length (cm) of lime seedlings following treatment with Trichoderma and coconut water

Trichoderma sp.	Coconut water concentration				Average
	A0	A1	A2	A3	
T0	9.7	7.8	8.9	9.4	9.0[b]
T1	11.9	8.9	9.5	9.8	10.1[ab]
T2	11.6	10.3	10.3	11.5	10.9[a]
Average	11.0[a]	9.0[b]	9.5[ab]	10.2[ab]	
HSD (A)	1.9				
HSD (T)	1.5				

Note: Values in the same row (a, b, and c) and column (x, y and z) that are followed by different letters indicate significant differences at the 0.05% level according to the HSD test.

The results from the 5% HSD test in Table 2 indicate that the application of 50 g Trichoderma per plant (T2) significantly enhances shoot length, achieving an average of 10.9 cm compared to 9.0 cm in the control without Trichoderma (T0). This suggests that Trichoderma positively influences shoot growth, likely through mechanisms such as improved nutrient uptake, enhanced root development, or induced systemic resistance that promotes overall plant vigor (Andrzejak & Janowska, 2022). The significant difference between T2 and T0 confirms the efficacy of Trichoderma at this application rate in stimulating shoot elongation.

Similarly, the coconut water immersion treatments reveal that 100% coconut water (A0) yields the longest shoots at 11.0 cm, significantly outperforming the 15% coconut water treatment (A1), which results in the shortest average shoot length of 9.0 cm. This implies that the concentration of coconut water plays a critical role in promoting shoot growth, possibly due to the higher availability of growth-promoting substances such as cytokinins, vitamins, and minerals in the undiluted coconut water. The lower shoot length observed in the 15% treatment may reflect insufficient levels of these bioactive compounds to stimulate optimal growth (Andrzejak & Janowska, 2022).

Together, these findings highlight the importance of both Trichoderma application and coconut water concentration as influential factors for maximizing shoot length. The synergistic potential of combining effective Trichoderma doses with high-concentration coconut water immersion could be explored further to optimize growth outcomes(Rubayet & Hossain, 2025; Supyanto & Argus, 2025). Additionally, the significant differences underscore the need to carefully consider treatment levels to achieve the best physiological responses in plants (Rubayet & Hossain, 2025).

4. Number of leaves

The observational results concerning the number of leaves on lime seedling cuttings, following the application of Trichoderma and coconut water, were analyzed. The analysis of variance indicated that the application of Trichoderma and coconut water significantly affected the number of leaves on lime seedlings. However, the interaction between Trichoderma and coconut water application did not significantly influence the number of leaves on lime seedlings.

Table 3. Average number of leaves of lime seedlings after application of Trichoderma and coconut water

Trichoderma sp.	Coconut water concentration				Average
	A0	A1	A2	A3	
T0	7.7	6.0	7.0	7.0	6.9 [b]
T1	10.0	6.3	7.0	7.7	7.8 [ab]
T2	10.3	8.0	8.3	9.7	9.1 [a]

Average	9.3[a]	6.7 [b]	7.4 [ab]	8.1 [ab]
HSD (A)	2.2			
HSD (T)	1.8			

Note: Numbers followed by distinct letters within the same row (a, b, and c) and column (x, y and z) exhibit significant differences at the 0.05% level according to the HSD test.

The results from the 5% HSD test in Table 3 indicate that the application of 50 g Trichoderma per plant (T2) significantly enhances the average number of leaves compared to the control treatment without Trichoderma (T0). This suggests that Trichoderma acts as a beneficial biostimulant, likely promoting plant growth through mechanisms such as improved nutrient uptake, enhanced root development, or induced systemic resistance against pathogens. The notable difference between T2 and T0 highlights the positive impact of Trichoderma on leaf production, which is a critical indicator of vegetative vigor and overall plant health (Khan et al., 2023; Zhu et al., 2022).

The coconut water immersion treatments exhibit a distinct dose-dependent impact on leaf number. The treatment with 100% coconut water (A0) resulted in the highest average leaf count, significantly surpassing the 15% coconut water treatment (A1). Coconut water comprises various growth-promoting substances, such as cytokinins, vitamins, and minerals, which likely facilitate enhanced cell division and elongation, thereby increasing leaf formation. The reduced leaf count observed in the 15% treatment indicates that dilution decreases the availability of these bioactive compounds, thereby diminishing their stimulatory effect (Noor et al., 2026).

Together, these findings underscore the synergistic potential of biological and natural growth enhancers in optimizing plant development. The significant differences in leaf number across treatments indicate that both Trichoderma application and coconut water immersion can be strategically used to improve vegetative growth parameters (Chagas Junior et al., 2022). Future research could explore the combined effects of these treatments on other growth metrics, such as biomass accumulation and photosynthetic efficiency, to fully elucidate their agronomic value (Anhar et al., 2020; Chen et al., 2021).

5. Percentage of success

The success rate of lime seedling cuttings was lowest in treatments T0A1 and T1A1. Treatments without Trichoderma and 15% coconut water immersion, and with 25g/plant Trichoderma and 15% coconut water immersion, had a success rate of 53.33%. Data are shown in the graph in Figure 2 below:

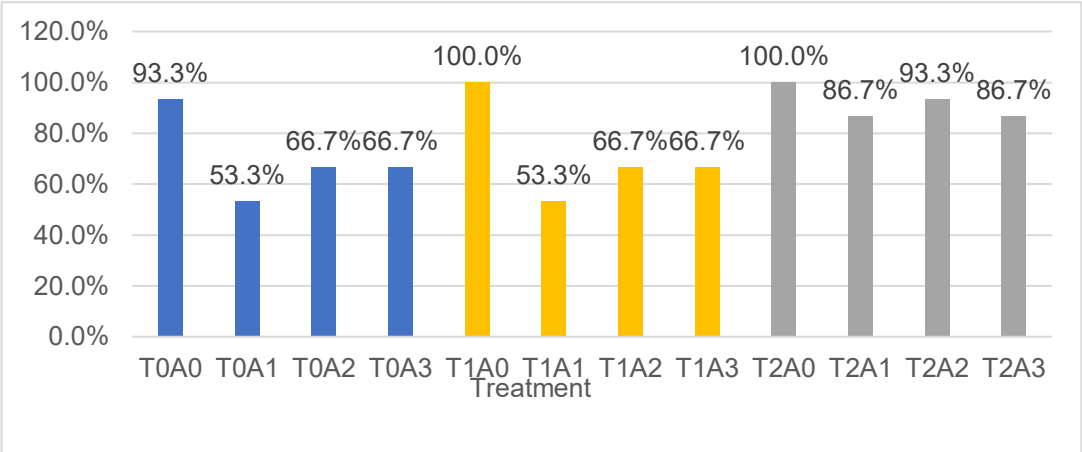

Figure 2. Diagram of the percentage of success of lime seedling cuttings at 12 WAP.

The results indicate a clear interaction effect between trichoderma application and coconut water concentration on the success rate of cuttings. The lowest success rate of 53% observed in treatments without trichoderma and with low (15%) coconut water immersion (T0A1), as well as with 25 g trichoderma combined with 15% coconut water (T1A1), suggests that insufficient nutrient or bioactive compound availability in the diluted coconut water may limit rooting and growth stimulation despite the presence of trichoderma in the latter case (Ginting et al., 2025; Indriyanti et al., 2020).

Conversely, the highest success rate of 100% in treatments with 25 g and 50 g of trichoderma combined with 100% coconut water immersion (T1A0 and T2A0) demonstrates a synergistic effect where

the full-strength coconut water likely provides optimal levels of growth-promoting hormones, vitamins, and minerals that enhance the beneficial action of trichoderma. Trichoderma, known for its role in improving plant health through pathogen suppression and growth promotion, appears most effective when paired with the rich biochemical environment of undiluted coconut water, facilitating better root initiation and establishment at 2 weeks after planting (WAP) (Ayyandurai et al., 2024; Maurya et al., 2024).

This suggests that both the concentration of coconut water and the dosage of trichoderma are critical factors influencing cutting success. The results highlight the importance of optimizing both parameters to maximize propagation efficiency. Lower coconut water concentrations may dilute essential growth factors, reducing the efficacy of trichoderma, while higher concentrations enhance the microbial and hormonal support needed for cutting survival and growth (Qomariyah et al., 2025).

Further investigation into the biochemical interactions between trichoderma metabolites and coconut water components could elucidate the mechanisms underlying this enhanced success rate. Additionally, assessing the root morphology and physiological parameters in these treatments would provide deeper insights into how these factors contribute to cutting vigor and establishment.

CONCLUSIONS

The application of Trichoderma at a rate of 50 g per plant yielded optimal outcomes, characterized by shoot emergence occurring 7.9 days post-planting, a shoot length of 10.9 cm, and the development of 9.1 leaves. Similarly, immersing lime cuttings in a 100% coconut water solution resulted in superior effects, with shoot emergence at 7.8 days, a shoot length of 11.0 cm, and 9.3 leaves. Although the interaction between Trichoderma and coconut water was not statistically significant, the combined treatment of 50 g of Trichoderma per plant with a 100% coconut water concentration exhibited a tendency towards a positive impact, evidenced by shoot emergence at 6.7 days post-planting, the production of 1.7 shoots, a shoot length of 11.6 cm, 10.3 leaves, and 100% growth.

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