Agronomy Research **24**(S1), 225–236, 2026
https://doi.org/10.15159/AR.26.047

Early morphogenesis of kopyor coconut (*Cocos nucifera* L.) embryos *in vitro* as affected by light conditions and thidiazuron concentration

S. Susiyanti[1,5,*], W.S. Hamidah[1], A.A. Fatmawaty[1], Z. Millah[1], Kartina A.M.,[1] M. Mirajiani[2], S. Abdullah[3] and S. Sjaifuddin[4]

[1]Department of Agroecotechnology, Faculty of Agriculture, Universitas Sultan Ageng Tirtayasa, Serang, Banten Province, Indonesia
[2]Department of Agribusiness, Faculty of Agriculture, Universitas Sultan Ageng Tirtayasa, Serang, Banten Province, Indonesia
[3]Faculty of Plantation and Agrotechnology, Universiti Teknologi MARA, Malaysia
[4]Department of Environmental Studies, Universitas Sultan Ageng Tirtayasa, Serang, Banten Province, Indonesia
[5]Center of Excellent for Food Security, Universitas Sultan Ageng Tirtayasa, Serang, Banten Province, Indonesia
[*]Correspondence: susiyanti@untirta.ac.id

Received: December 31st, 2025; Accepted: May 7th, 2026; Published: June 10th, 2026

Abstract. Kopyor coconut (*Cocos nucifera* L.) is a high-value coconut variant characterized by a naturally mutated endosperm; however, its propagation is constrained by low seed viability and inconsistent germination. *In vitro* culture offers a promising approach for large-scale propagation. This study evaluated the effects of light conditions and thidiazuron (TDZ) concentrations on the early morphogenesis of kopyor coconut embryos cultured *in vitro*. A split-plot design based on a completely randomized design was applied, with light conditions (dark and continuous light) as the main plot and TDZ concentrations (0, 0.25, 0.50, 0.75, and 1.00 mg L^{-1}) as subplots. The results showed that light conditions did not significantly affect explant height, shoot number and root number, or root length ($p > 0.05$), although plumule formation occurred exclusively under light conditions. In contrast, TDZ significantly influenced explant height ($p < 0.01$), while its effects on other parameters were not significant. The application of 0.25 mg L^{-1} TDZ produced the most favorable response, with explant height reaching 3.00 ± 0.14 cmunder light and 2.60 ± 0.17 cmunder dark conditions, accompanied by 0.95 shoots and 0.86 roots per explant. No significant interaction between light conditions and TDZ was detected. These findings indicate that early morphogenesis of kopyor coconut embryos is primarily regulated by endogenous hormonal balance, with low TDZ concentrations optimizing growth responses, while light plays a critical role in organ differentiation, particularly in plumule formation. The results provide a basis for improving *in vitro* propagation protocols for this economically important coconut variant.

Key words: *Cocos nucifera* L., kopyor coconut, morphogenesis, light conditions, thidiazuron, *in vitro*.

INTRODUCTION

Indonesia is widely recognized as a megadiversity country with tropical agroecological conditions that strongly support perennial crops, particularly coconut (*Cocos nucifera* L.), which plays an important economic and ecological role due to the extensive utilization of its plant parts. Among coconut variants, kopyor coconut represents a unique and high-value type resulting from a natural mutation in the endosperm. This mutation produces a soft, friable endosperm with a distinctive aroma, attributes that significantly increase its commercial value and consumer demand (Mu et al., 2024).

Despite its high economic potential, kopyor coconut production in Indonesia remains limited, particularly in key regions such as Pati, Central Java, due to the scarcity of viable planting materials and the instability of seedling supply. Low seed viability and the unpredictable outcomes of conventional propagation often result in poor germination and inconsistent plant performance, thereby constraining large-scale cultivation and commercialization (Long et al., 2022). Therefore, alternative propagation strategies are required to produce uniform and reliable planting materials.

In vitro culture has been widely applied as an alternative propagation approach for perennial crops, including coconut, due to its advantages in rapid multiplication, genetic uniformity, and production of pathogen-free plantlets (Nguyen et al., 2016; Kalaipandian et al., 2021). However, coconut is considered a recalcitrant species in tissue culture, with morphogenic responses strongly influenced by basal medium composition, plant growth regulators (PGRs), and environmental conditions. Y3 medium is commonly used as the standard basal medium for coconut tissue culture and is typically supplemented with carbohydrates, vitamins, and growth regulators to compensate for the limited photosynthetic capacity of *in vitro* explants (Zhang et al., 2023; Pokimica et al., 2024).

Plant growth regulators play a central role in controlling *in vitro* morphogenesis. Cytokinins generally promote shoot development, whereas auxins are associated with root and callus formation (Zhang et al., 2023). Thidiazuron (TDZ), a synthetic cytokinin, has been widely reported to be highly effective in stimulating shoot proliferation in coconut and other woody species. However, its effects are strongly dependent on concentration, explant type, and culture conditions (Wilms et al., 2021; Kong et al., 2025). High TDZ concentrations may inhibit growth or induce abnormal morphogenesis, highlighting the need for precise optimization. In kopyor coconut, available studies remain limited and primarily focus on embryogenic callus induction using auxin-cytokinin combinations, while information on optimal TDZ concentrations for early morphogenesis is still lacking (Chandrakala et al., 2019).

In addition to PGRs, light conditions are an important environmental factor influencing *in vitro* morphogenesis through their effects on chlorophyll synthesis, cell elongation, and organ differentiation. Although light regimes have been reported to enhance plantlet growth and acclimatization (Bett et al., 2019; Mu et al., 2025), information regarding the role of light during early developmental stages of kopyor coconut embryos remains limited and inconsistent, particularly in relation to cytokinin-mediated responses.

Therefore, this study aimed to evaluate the effects of light conditions (light and dark) and different TDZ concentrations (0, 0.25, 0.50, 0.75, and 1.00 mg L^{-1}) on early morphogenesis of kopyor coconut embryos cultured *in vitro*. This study addresses the existing gap in understanding the interaction between hormonal regulation and light-mediated developmental processes and provides a basis for optimizing efficient and standardized propagation protocols for this high-value coconut variant.

MATERIALS AND METHODS

Experimental site and duration

The experiment was conducted from September to December 2025 at the Laboratory of Physiology and Biotechnology, Universitas Sultan Ageng Tirtayasa, Indonesia, under controlled laboratory conditions.

Plant materials and experimental design

Zygotic embryos of kopyor coconut (*Cocos nucifera* L.) obtained from Pati, Central Java, Indonesia, were used as explants. The embryos were derived from fruits characterized by brown-colored exocarp and yellow petiole, which are typical morphological traits of the selected kopyor type. The experiment was arranged in a split-plot completely randomized design (CRD) with two factors. Light condition was assigned as the main plot, consisting of complete darkness (C_0) and continuous light (C_1). TDZ concentration was designated as the subplot factor, with five levels: 0, 0.25, 0.50, 0.75, and 1.00 mg L^{-1}. Each treatment combination was replicated three times, with each experimental unit consisting of one explant (one embryo), resulting in a total of 30 experimental units in the experiment.

Culture media preparation

The basal medium used in this study was Y3 medium following Eeuwens formulation. The medium was supplemented with 15 g L^{-1} sucrose, 1 g L^{-1} activated charcoal, 50 mL L^{-1} coconut water, 7 g L^{-1} agar, and 0.5 mL L^{-1} Plant Preservative Mixture (PPM). TDZ was added according to the respective treatment concentrations. The pH of the medium was adjusted to 5.6 prior to sterilization, and all media were autoclaved at 121 °C for 15 min.

Sterilization and inoculation procedures

All instruments and culture vessels were sterilized by autoclaving. Inoculation was performed aseptically in a laminar airflow cabinet sterilized with ultraviolet light for 30 min. Instruments were disinfected with 70% ethanol and flamed during inoculation. After inoculation, culture vessels were sealed to prevent contamination.

Culture conditions and treatments

Cultures were maintained at 22–26 °C for eight weeks. Two light conditions were applied: complete darkness (0 lux) and continuous illumination at approximately 2,000 lux provided by cool-white fluorescent lamps (2×40 W) positioned approximately 30 cmabove the culture shelves.

Data collection

Observations were conducted until week 8. Parameters recorded included explant height (cm), number of shoots per explant, number of roots per explant, and root length (cm). Additional parameters included the percentage of rooted explants and percentage of plumule emergence.

Statistical analysis

Data were subjected to analysis of variance (ANOVA) at a 5% significance level. When significant differences were detected, Duncan's Multiple Range Test (DMRT) was applied. Statistical analysis was performed using DSAASTAT version 1.101.

RESULTS AND DISCUSSION

The effects of light conditions and TDZ on *in vitro* early shoot development of kopyor coconut were evaluated using multiple growth parameters. Analysis of variance (Table 1) showed that light conditions did not significantly affect explant height, shoot number and root number, or root length. In contrast, TDZ concentration had a highly significant effect ($p < 0.01$) on explant height, while its effects on other parameters were not significant.

Table 1. Analysis of variance (ANOVA) summary of the effects of light conditions and TDZ on early morphogenesis of kopyor coconut (*Cocos nucifera* L.) embryos cultured *in vitro*

Parameter	Light condition (C)	TDZ concentration (T)	Interaction (C × T)	CV main plot (%)	CV subplot (%)
Explant height	ns	**	ns	14.31	20.02
Number of shoots	ns	ns	ns	10.49	16.58
Number of roots	ns	ns	ns	17.79	16.13
Root length	ns	ns	ns	6.20	10.91

Note: ns = not significant ($p > 0.05$); ** = highly significant at $p \leq 0.01$; CV = coefficient of variation.

No significant interaction between light conditions and TDZ concentration was detected, suggesting that TDZ responses were largely independent of light conditions. Coefficients of variation ranging from 6.20% to 20.02% indicate acceptable experimental precision.

Although no significant differences in explant height were observed between light and dark conditions ($p > 0.05$), plumule formation occurred only under light conditions and was absent in cultures maintained in darkness.

Explant height

Explant height increased gradually over the eight-week culture period. Light conditions had no significant effect on explant height, whereas TDZ concentration had a highly significant effect ($p < 0.01$), with no significant interaction between the two factors. The detailed effects of treatments on explant height are presented in Table 2.

The slightly greater explant height observed under dark conditions (2.52 cm) compared to light conditions (2.45 cm) may be associated with etiolation, a typical response to the absence of light that promotes elongation (Armarego-Marriott et al., 2020; Huang et al., 2025). However, this difference was not statistically significant ($p > 0.05$).

Table 2. Effect of light conditions and TDZ on explant height of kopyor coconut (*Cocos nucifera* L.) embryos cultured *in vitro* (cm)

Light condition	TDZ Concentration (mg L^{-1})					Mean
	0.00 (T_0)	0.25 (T_1)	0.50 (T_2)	0.75 (T_3)	1.00 (T_4)	
Dark (C_0)	1.81 ± 0.27	2.60 ± 0.17	2.69 ± 0.52	2.82 ± 0.06	3.11 ± 0.33	2.52
Light (C_1)	1.66 ± 0.33	3.00 ± 0.14	2.55 ± 0.21	2.40 ± 0.08	2.66 ± 0.16	2.45
Mean	1.73^{c}	2.80^{a}	2.62^{b}	2.61^{ab}	2.88^{a}	2.49

Note: Values are expressed as mean ± SEM. Means followed by different letters within the TDZ factor differ significantly according to DMRT at $p \leq 0.05$.

Although light is known to regulate photomorphogenesis through processes such as cell differentiation, chloroplast development, and organ formation (Zhang et al., 2024¸ Wu et al., 2025), the comparable explant height observed in this study suggests that elongation under dark conditions may compensate for reduced photomorphogenic development.

TDZ at 0.25 and 1.00 mg L^{-1} produced the highest explant height (2.80–2.88 cm). DMRT analysis confirmed that 0.25 mg L^{-1} TDZ resulted in explant height statistically comparable to higher concentrations but significantly greater than the control and 0.50 mg L^{-1} treatments. This indicates that low TDZ concentrations are sufficient to promote shoot elongation without inducing inhibitory effects.

TDZ-induced morphogenesis is associated with complex hormonal regulation pathways (Erland et al., 2020; Ghosh et al., 2018). The response of explant height to TDZ was not linear, as similar growth responses were observed at both 0.25 and 1.00 mg L^{-1}. This pattern may reflect interactions between exogenous TDZ and endogenous hormones present in kopyor coconut embryos. Zygotic embryos of coconut are known to contain relatively high endogenous cytokinin levels, and TDZ may further modulate the cytokinin-auxin balance, resulting in a non-linear growth response (Kalaipandian et al., 2021). This finding is consistent with previous reports that TDZ influences plant development through its cytokinin-like activity and regulation of hormonal crosstalk (Ali et al., 2022; Li et al., 2022; Faisal et al., 2023).

Number of shoots per explant

Shoot initiation was observed during the culture period, although its timing was not quantitatively assessed. Overall, shoot formation was not significantly influenced by light conditions, TDZ concentration, or their interaction ($p > 0.05$). The mean number of shoots remained low, ranging from 0.81 to 0.95 shoots per explant across treatments. The response of shoot number across treatments is summarized in Table 3.

The limited shoot induction observed in this study may be attributed to the balance of endogenous auxin and cytokinin (Van Gelderen et al., 2021; Kong et al., 2025), as well as the type of explant used. Zygotic embryos are developmentally programmed to produce a single primary shoot during germination, which inherently limits the capacity for multiple shoot formation under *in vitro* conditions (Nguyen et al., 2016). In addition, the use of zygotic embryos as explants may further contribute to the low shoot formation observed, as these tissues are primarily oriented toward germination rather than proliferation, thereby limiting their morphogenic potential for multiple shoot induction.

Table 3. Effect of light conditions and TDZ on shoot number of kopyor coconut (*Cocos nucifera* L.) embryos cultured *in vitro*

Light condition	TDZ (mg L^{-1})					Mean
	0.00 (T_0)	0.25 (T_1)	0.50 (T_2)	0.75 (T_3)	1.00 (T_4)	
Dark (C_0)	0.81 ± 0.10	0.90 ± 0.10	0.90 ± 0.10	1.00 ± 0.00	0.90 ± 0.10	0.90
Light (C_1)	0.81 ± 0.10	1.00 ± 0.00	0.90 ± 0.10	0.71 ± 0.00	0.81 ± 0.10	0.85
Mean	0.81	0.95	0.90	0.86	0.86	0.87

Note: Values are expressed as mean ± SEM. No significant differences were observed among treatments according to analysis of variance (ANOVA) at $p > 0.05$.

A similar pattern was observed in root-related parameters (Table 4). Overall, these findings indicate that the morphogenic response of kopyor coconut embryos was low and not strongly influenced by the applied treatments.

Table 4. Effect of light conditions and TDZ on number of roots of kopyor coconut (*Cocos nucifera* L.) embryos cultured *in vitro*

Light condition	TDZ (mg L^{-1})					Mean
	0.00 (T_0)	0.25 (T_1)	0.50 (T_2)	0.75 (T_3)	1.00 (T_4)	
Dark (C_0)	0.71 ± 0.00	0.81 ± 0.10	0.81 ± 0.00	0.90 ± 0.10	0.81 ± 0.10	0.81
Light (C_1)	0.71 ± 0.00	0.90 ± 0.10	0.81 ± 0.10	0.71 ± 0.00	0.71 ± 0.00	0.77
Mean	0.71	0.86	0.81	0.81	0.76	0.79

Note: Values are expressed as mean ± SEM. No significant differences were observed among treatments according to analysis of variance (ANOVA) at $p > 0.05$.

Number of roots per explant

The number of roots per explant remained low (<1 root) across all treatments, and statistical analysis showed no significant effects of light conditions, TDZ concentration, or their interaction ($p > 0.05$). Numerically, rooting was slightly higher under dark conditions (mean = 0.81) than under light conditions (0.77), with the highest values observed at 0.75 mg L^{-1} in darkness (0.90) and 0.25 mg L^{-1} under light conditions (0.90). However, these differences were not statistically significant, indicating that light conditions had a limited influence on early rhizogenesis.

The limited rooting response observed in this study suggests that rhizogenesis in coconut is not strongly promoted by cytokinin. TDZ is known to stimulate shoot meristem activity but may inhibit root initiation at higher concentrations (Kalaipandian et al., 2021). Root development in palms is generally auxin-dependent and often requires exogenous auxin, such as indole-3-butyric acid (IBA) or naphthaleneacetic acid (NAA), as well as multi-stage culture systems to achieve effective rooting (Chandrakala et al., 2019).

In addition, the use of zygotic embryos as explants may have contributed to the low rooting response observed. Zygotic embryos are developmentally oriented toward germination rather than organ proliferation, and their endogenous hormonal balance is typically optimized for primary root formation rather than multiple root induction. This intrinsic developmental constraint may limit their responsiveness to exogenous treatments aimed at enhancing rhizogenesis.

Therefore, the low rooting response may be attributed to a combination of limited endogenous auxin levels, the absence of auxin supplementation, and the inherent recalcitrant nature of monocot embryos (Martins et al., 2024).

Root length

Root length was relatively uniform across treatments, and analysis of variance showed no significant effects of light conditions or TDZ concentration ($p > 0.05$), indicating limited variation in root elongation. Root length tended to decrease with increasing TDZ concentration; although this trend was not statistically significant. Root length responses under different treatments are shown in Table 5.

Table 5. Effect of light conditions and TDZ on root length of kopyor coconut (*Cocos nucifera* L.) embryos cultured *in vitro* (cm)

Light condition	TDZ (mg L^{-1})					Mean
	0.00 (T_0)	0.25 (T_1)	0.50 (T_2)	0.75 (T_3)	1.00 (T_4)	
Dark (C_0)	1.10 ± 0.01	1.05 ± 0.02	1.07 ± 0.01	0.93 ± 0.05	1.08 ± 0.01	1.05
Light (C_1)	1.10 ± 0.02	1.09 ± 0.05	1.29 ± 0.17	1.10 ± 0.00	1.10 ± 0.00	1.14
Mean	1.10	1.07	1.18	1.01	1.09	1.09

Note: Values are expressed as mean ± SEM. No significant differences were observed among treatments according to analysis of variance (ANOVA) at $p > 0.05$.

The regulatory role of auxin in root formation has been widely reported (Du & Scheres, 2018). Root development occurred in the absence of exogenous auxin, suggesting that endogenous hormonal activity within the embryo was sufficient to support root initiation, potentially enhanced by growth-promoting compounds present in coconut water (Chandrakala et al., 2019; Martins et al., 2024). In palms, root formation is generally auxin-dependent and often requires exogenous auxin supplementation for optimal development (Kalaipandian et al., 2021).

The use of zygotic embryos as explants may have contributed to the observed root elongation pattern, as these tissues are developmentally oriented toward primary root establishment during germination rather than extensive root proliferation (Armenta-Medina et al., 2020; Venglat et al., 2025), which may limit their responsiveness to exogenous treatments (Tian et al., 2020).

Following shoot development, newly formed shoot tissues may have contributed additional endogenous growth regulators that supported subsequent root elongation. Although light conditions can influence auxin sensitivity and root development, these effects are often species-specific and not always significant (Yun et al., 2023). Overall, the results indicate that root elongation in kopyor coconut embryos was primarily regulated by endogenous hormonal balance rather than by light or TDZ treatments.

Root formation was observed at the basal region of the explant under both light and dark conditions at 0.25 mg L^{-1} TDZ (Fig. 1). The roots appeared as whitish, elongated structures emerging from compact root primordia, indicating active rhizogenesis. These observations confirm that the explants were capable of initiating and sustaining root growth under the applied treatment, although the overall rooting response remained limited. Consistent with the quantitative results, rhizogenesis in kopyor coconut embryos appears to be primarily regulated by endogenous hormonal balance rather than by TDZ concentration or light conditions (Pasternak & Steinmacher, 2024; Indrachapa et al., 2025).

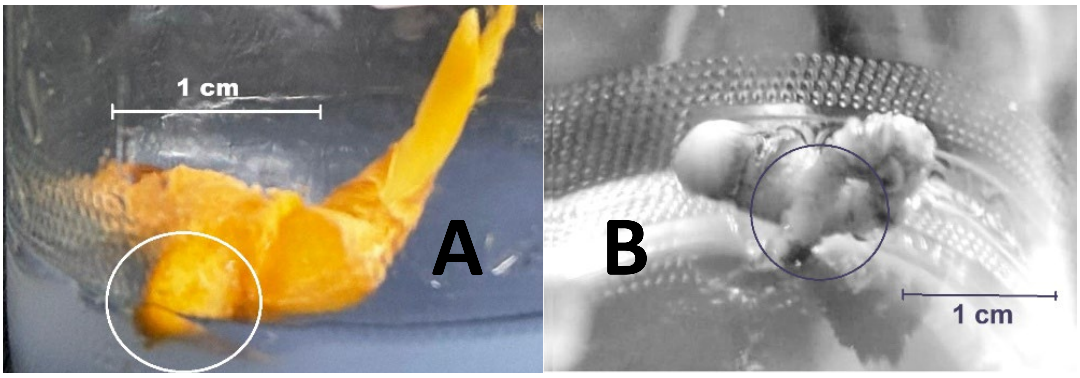

Figure 1. Root formation in kopyor coconut explants cultured under 0.25 mg L^{-1} TDZ under light (A) and dark conditions (B). Root primordia (circled) developed at the basal region and elongated into visible roots, indicating active rhizogenesis. Scale bar = 1 cm.

Percentage of rooted explants

Based on root formation observations, 8 out of 30 explants developed roots, resulting in an overall rooting percentage of 26.67%. Rooting was numerically higher under dark conditions (33.33%) than under light conditions (20%). Under darkness, root formation was observed only in TDZ-treated explants (C_0T_1–C_0T_4), whereas under light conditions it was limited to treatments C_1T_1 and C_1T_2.

The higher rooting percentage observed under dark conditions may be associated with reduced light exposure, which can enhance endogenous auxin activity by limiting its degradation (Van Gelderen et al., 2021). Although fewer explants formed roots under light conditions, those that did tended to produce longer roots, suggesting that light may influence root elongation rather than initiation. However, these patterns were not statistically significant and should therefore be interpreted with caution.

In addition, the use of zygotic embryos as explants may have contributed to the low rooting frequency observed. These embryos are developmentally oriented toward germination rather than organ proliferation, which may limit their responsiveness to treatments aimed at enhancing rhizogenesis.

Overall, these findings suggest that root development in kopyor coconut embryos was primarily regulated by endogenous hormonal balance, particularly auxin activity, while the effects of TDZ and light conditions appeared to be limited under the experimental conditions (Kim et al., 2023). Further studies incorporating exogenous auxin supplementation are required to better elucidate the mechanisms underlying root initiation and development. Similar observations have been reported in other *in vitro* systems, where endogenous hormonal activity was sufficient to support rhizogenesis (Faisal et al., 2023).

Percentage of plumule development

Although no significant differences in explant height were observed between light and dark conditions ($p > 0.05$), plumule formation occurred exclusively under light conditions. This indicates that light had a limited effect on elongation but played a critical role in photomorphogenesis, particularly in regulating organ differentiation and chlorophyll development during early embryo growth (Wu et al., 2025; Manghwar et al., 2026).

Plumules, characterized by leaf-like structures (Fig. 2), were observed only under light conditions, with a maximum frequency of 10%, particularly at low TDZ concentrations (C_1T_0, C_1T_1, and C_1T_2). No plumule formation occurred in explants cultured in darkness, highlighting the importance of light in leaf primordia differentiation (Van Gelderen et al., 2018; Kong et al., 2025).

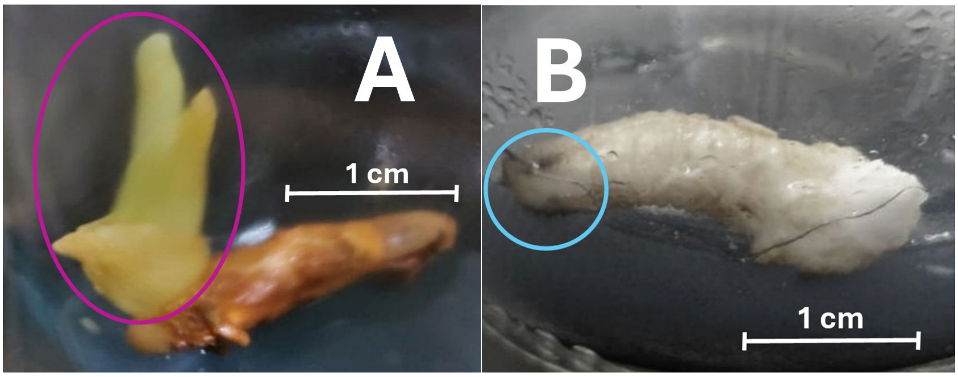

Figure 2. Plumule formation in kopyor coconut embryos cultured under light conditions with 0.5 mg L^{-1} TDZ (A), showing a well-developed shoot structure, and the absence of visible plumule under dark conditions with 0.5 mg L^{-1} TDZ (B). Scale bar = 1 cm.

High TDZ concentrations did not promote plumule formation, which is consistent with previous findings that excessive cytokinin levels may inhibit shoot differentiation (Akbar et al., 2018). In addition, the use of zygotic embryos as explants may have influenced plumule initiation, as their developmental stage is primarily oriented toward germination rather than active shoot proliferation, potentially limiting morphogenic competence (Sun et al., 2025).

Plumule formation was observed only under light conditions (Fig. 2, A), where a distinct, elongated shoot structure developed. In contrast, no visible plumule was formed under dark conditions (Fig. 2, B), and the explants remained morphologically undifferentiated. These observations confirm that light is essential for plumule initiation and shoot differentiation in kopyor coconut embryos.

CONCLUSION

Light conditions did not significantly affect quantitative growth parameters, including explant height, shoot number and root number, and root length of kopyor coconut cultured *in vitro*. However, plumule development occurred exclusively under light conditions, indicating that light plays a critical role in early organ differentiation despite its limited effect on overall growth.

In contrast, TDZ concentration significantly influenced explant height, with 0.25 mg L^{-1} producing the most favorable response. Although similar trends were observed for shoot and root development, these effects were not statistically significant, and no significant interaction between light conditions and TDZ concentration was detected.

These findings demonstrate that early morphogenesis of kopyor coconut embryos is primarily regulated by endogenous hormonal balance, with light acting as a key factor in developmental differentiation rather than growth promotion. This study provides insight into the early developmental constraints of kopyor coconut in vitro and highlights the need to optimize hormonal combinations, particularly through the inclusion of exogenous auxin, to enhance shoot and root development. Future research should further explore the interaction between endogenous hormones and light signaling to improve propagation efficiency.

REFERENCES

Ali, H.M., Khan, T., Khan, M.A. & Ullah, N. 2022. The multipotent thidiazuron: A mechanistic overview of its roles in callogenesis and other plant cultures *in vitro*. *Biotechnology and Applied Biochemistry* **69**(6), 2624–2640. https://doi.org/10.1002/bab.2311

Armarego-Marriott, T., Sandoval-Ibañez, O. & Kowalewska, Ł. 2020. Beyond the darkness: Recent lessons from etiolation and de-etiolation studies. *Journal of Experimental Botany* **71**(21), 6709–6721. https://doi.org/10.1093/jxb/eraa356

Armenta-Medina, A., Gillmor, C.S., Gao, P., Mora-Macias, J., Kochian, L.V., Xiang, D. & Datla, R. 2020. Developmental and genomic architecture of plant embryogenesis: From model plant to crops. *Plant Communications* **2**(1), 100136. https://doi.org/10.1016/j.xplc.2020.100136

Bett, C.C., Mweu, C.M. & Nyende, A.B. 2019. In vitro regeneration of coconut (*Cocos nucifera* L.) through indirect somatic embryogenesis in Kenya. *African Journal of Biotechnology* **18**(32), 1113–1122. https://doi.org/10.5897/AJB2019.16867

Chandrakala, D., Renuka, R. & Sushmitha, D.S. 2019. Influence of 2, 4-d and TDZ on direct organogenesis in coconut var. east coast tall. *Int. J. Chem Stud.* **7**(3). 4111–4115.

Du, Y. & Scheres, B. 2018. Lateral root formation and the multiple roles of auxin. *Journal of Experimental Botany* **69**(2), 155–167. https://doi.org/10.1093/jxb/erx223

Erland, L.A.E., Giebelhaus, R.T., Victor, J.M.R., Murch, S.J. & Saxena, P.K. 2020. The morphoregulatory role of thidiazuron: Metabolomics-guided hypothesis generation for mechanisms of activity. *Frontiers in Plant Science* **11**, 599496. https://doi.org/10.3389/fpls.2020.599496

Faisal, M., Qahtan, A.A. & Alatar, A.A. 2023. Thidiazuron-induced in vitro plant regeneration, phenolic contents, antioxidant potential, GC-MS profiles, and nuclear genome stability of *Plectranthus amboinicus* (Lour.) Spreng. *Horticulturae* **9**(2), 277. https://doi.org/10.3390/horticulturae9020277

Ghosh, A., Igamberdiev, A.U. & Debnath, S.C. 2018. Thidiazuron-induced somatic embryogenesis and changes of antioxidant properties in tissue cultures of half-high blueberry plants. *Scientific Reports* **8**, 16978. https://doi.org/10.1038/s41598-018-35373-7

Huang, Z., Liu, Q., Zou, X., Zhu, L., Ma, X. & Huang, R. 2025. Morpho-physiological responses during dark-induced leaf senescence in *Cunninghamia lanceolata* seedlings. *Forests* **16**(9), 1372. https://doi.org/10.3390/f16091372

Indrachapa, M.T.N., Balasuriya, B.M.I.K., Jayarathna, S.P.N.C., Vidhanaarachchi, V.R.M. & Mu, Z. 2025. Optimizing pulse treatments for enhanced in vitro rooting in coconut micropropagation. *Technology in Horticulture* **5**, e027. https://doi.org/10.48130/tihort-0025-0019

Kalaipandian, S., Mu, Z., Kong, E.Y.Y., Biddle, J., Cave, R., Bazrafshan, A., Wijayabandara, K., Beveridge, F.C., Nguyen, Q. & Adkins, S.W. 2021. Cloning coconut via somatic embryogenesis: A review of the current status and future prospects. *Plants* **10**(10), 2050. https://doi.org/10.3390/plants10102050

Kim, S.-H., Zebro, M., Jang, D.-C., Sim, J.-E., Park, H.-K., Kim, K.-Y., Bae, H.-M., Tilahun, S. & Park, S.-M. 2023. Optimization of plant growth regulators for in vitro mass propagation of a disease-free 'Shine Muscat' grapevine cultivar. *Current Issues in Molecular Biology* **45**(10), 487. https://doi.org/10.3390/cimb45100487

Kong, E.Y.Y., Biddle, J.M., Sisunandar, S., Kalaipandian, S., Bazrafshan, A., Mu, Z. & Adkins, S.W. 2025. Initiation of direct shoot organogenesis in coconut using immature inflorescence. *Plants* **14**(20), 3123. https://doi.org/10.3390/plants14203123

Li, F., Wu, Q., Liao, B., Yu, K., Huo, Y., Meng, L., Wang, S., Wang, B., Du, M., Tian, X. & Li, Z. 2022. Thidiazuron promotes leaf abscission by regulating the crosstalk between ethylene, auxin, and cytokinin in cotton. *International Journal of Molecular Sciences* **23**(5), 2696. https://doi.org/10.3390/ijms23052696

Long, Y., Yang, Y., Pan, G. & Shen, Y. 2022. New insights into tissue culture plant-regeneration mechanisms. *Frontiers in Plant Science* **13**, 926752. https://doi.org/10.3389/fpls.2022.926752

Manghwar, H., Hussain, A., Alam, I., Fan, S., Sun, G. & Liu, F. 2026. Brassinosteroids (BRs) and photomorphogenesis: Hormonal regulation of light-mediated plant development. *Environmental and Experimental Botany* **241**, 106298. https://doi.org/10.1016/j.envexpbot.2025.106298

Martins, J.P.R., Wawrzyniak, M.K., Kalemba, E.M., Ley-López, J.M., Sousa Lira, J.M. & Chmielarz, P. 2024. In vitro rooting of *Quercus robur*: Activated charcoal vs. exogenous auxin—A morphophysiological approach. *Plant Cell, Tissue and Organ Culture* **156**, 24–35. https://doi.org/10.1007/s11240-023-02532-4

Mu, Z., Li, Z., Nulu, N.P.C., Kalaipandian, S., Biddle, J.M., Bazrafshan, A., Kong, E. & Adkins, S.W. 2025. A photomixotrophic system to improve the growth of in vitro cultured seedlings of coconut (*Cocos nucifera* L.). *Horticulturae* **11**(3), 224. https://doi.org/10.3390/horticulturae11030224

Mu, Z., Yang, Z., Xu, H., Khongmaluan, M., Arikit, S., Tran, B.-M.T., Vidhanaarachchi, V.R. M., Sisunandar, S., Yang, S., Peng, H. & Luo, J. 2024. Prospects and challenges of elite coconut varieties in China: A case study of makapuno. *Tropical Plants* **3**, e029. https://doi.org/10.48130/tp-0024-0007

Nguyen, Q.T., Bandupriya, H.D.D., López Villalobos, A., Sisunandar, S., Foale, M. & Adkins, S.W. 2016. Biology, propagation, and utilization of elite coconut varieties. *Plant Physiology and Biochemistry* **109**, 579–589. https://doi.org/10.1016/j.plaphy.2016.10.007

Pasternak, T.P. & Steinmacher, D. 2024. Plant growth regulation in cell and tissue culture *in vitro*. *Plants* **13**(2), 327. https://doi.org/10.3390/plants13020327

Pokimica, N., Cosić, T., Uzelac, B., Ninković, S. & Raspor, M. 2024. Dissecting the roles of the cytokinin signaling network: The case of de novo shoot apical meristem formation. *Biomolecules* **14**(3), 381. https://doi.org/10.3390/biom14030381

Sun, J., Li, J., Zhao, M., Zheng, G., Di, B., Wang, J. & Hu, R. 2025. Successful establishment of somatic embryogenesis and shoot organogenesis systems in *Catalpa bungei*. *Plants* **14**(17), 2688. https://doi.org/10.3390/plants14172688

Tian, R., Paul, P., Joshi, S. & Perry, S.E. 2020. Genetic activity during early plant embryogenesis. *Biochemical Journal* **477**(19), 3743–3767. https://doi.org/10.1042/BCJ20190161

Van Gelderen, K., Kang, C. & Pierik, R. 2018. Light signaling, root development, and plasticity. *Plant Physiology* **176**(2), 1049–1060. https://doi.org/10.1104/pp.17.01207

Van Gelderen, K., Kang, C., Paalman, R., Keuskamp, D., Hayes, S. & Pierik, R. 2021. Regulation of lateral root development by shoot-sensed far-red light via HY5 movement to the root. *Frontiers in Plant Science* **12**, 660870. https://doi.org/10.3389/fpls.2021.660870

Venglat, P., Vijayan, P., Sharbel, T.F., Rahman, A. & Tanino, K. 2025. Phytomers, collet and founder cells: A 'universal' plant embryonic body plan. *Frontiers in Plant Science* **16**, 1521527. https://doi.org/10.3389/fpls.2025.1521527

Wilms, H., De Bièvre, D., Longin, K., Swennen, R., Rhee, J. & Panis, B. 2021. Development of the first axillary in vitro shoot multiplication protocol for coconut palms. *Scientific Reports* **11**, 18367. https://doi.org/10.1038/s41598-021-97888-1

Wu, W., Chen, L., Liang, R., Huang, S., Li, X., Huang, B., Luo, H., Zhang, M., Wang, X. & Zhu, H. 2025. The role of light in regulating plant growth, development and sugar metabolism: A review. *Frontiers in Plant Science* **15**, 1507628. https://doi.org/10.3389/fpls.2024.1507628

Yun, F., Liu, H., Deng, Y., Hou, X. & Liao, W. 2023. The role of light-regulated auxin signaling in root development. *International Journal of Molecular Sciences* **24**(6), 5253. https://doi.org/10.3390/ijms24065253

Zhang, K., Novak, O., Wei, Z., Gou, M., Zhang, X., Yu, Y., Setter, T. & Zhang, J. 2023. Spatially distributed cytokinins: Metabolism, signaling, and transport. *Cell Reports Physical Science* **4**(1), 101075. https://doi.org/10.1016/j.xcrp.2022.101075

Zhang, N., Wang, J., Liu, J., Zhang, P., Kudoyarova, G., Liu, C.J. & Zhang, K. 2024. Photoregulatory protein kinases fine-tune plant photomorphogenesis. *Developmental Cell* **59**(3), 232–245. https://doi.org/10.1016/j.devcel.2024.02.012