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Auxin, substrate and light interaction enhances rooting efficiency in *Taxus baccata* cuttings

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Review Article

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ABSTRACT

Taxus baccata L., an endangered woody species with poor rooting ability in stem cuttings, faces propagation challenges that limit its conservation and sustainable utilization. This study aimed to enhance rooting efficiency by investigating the combined effects of auxin type and concentration, substrate composition, and blue light exposure. A factorial experiment based on a completely randomized design with three replications was conducted, testing two auxins (NAA and IBA at 15 and 25 mg/L), two substrates (75% perlite + 25% peat moss and 100% perlite), and three blue light durations (2, 4, and 6 h/day). All main factors and their interactions markedly affected rooting percentage, callus formation, root biomass, survival, and rooting time. The treatment combining 25 mg/L NAA, 75% perlite + 25% peat moss, and 4 h/day of blue light produced the most favorable results, achieving the highest rooting and callus formation with the shortest rooting period. Blue light improved rooting performance by promoting auxin activity and stimulating physiological processes related to cell division and adventitious root initiation. Rooting time showed a negative relationship with callus formation, survival rate, and root biomass. These findings provide a mechanistic insight into auxin–substrate–light interactions and offer a practical protocol for efficient vegetative propagation and conservation of *Taxus baccata* L.

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1. Introduction

Vegetative propagation of *Taxus baccata* L. (European yew) is essential for both conservation and commercial purposes due to its slow natural regeneration and the high economic value of secondary metabolites such as taxol (Pipinis *et al*., 2023). However, stem cuttings of this species are notoriously difficult to root, with success influenced by factors including donor tree age, timing of cutting collection, and exogenous growth regulator application (Das & Jha, 2018a; Pipinis *et al*., 2023). Juvenile donor material consistently exhibits higher rooting capacity compared to mature tissue, underscoring the importance of physiological maturity in propagation efficiency (Bayraktar *et al*., 2018; Singh, 2021).

Auxins such as naphthaleneacetic acid (NAA) and indole-3-butyric acid (IBA) play pivotal roles in adventitious root induction. In this study, 25 mg/L NAA produced the highest rooting percentage, callus formation, and root biomass, consistent with findings in gymnosperm cuttings under optimized auxin treatments (Druege *et al*., 2019; Zhao, 2021; Martinez-Rodriguez *et al*., 2023). The superior performance of NAA over IBA may relate to its higher metabolic stability and sustained transport within plant tissues, leading to prolonged auxin signaling and enhanced root primordia initiation (Hernandez *et al*., 2020; Wang *et al*., 2021; Liu *et al*., 2022).

Substrate composition also significantly affects rooting outcomes. A mixture of 75% perlite and 25% peat moss outperformed pure perlite by providing improved moisture retention and aeration, critical factors for root growth and development (Gupta *et al*., 2020; Chen *et al*., 2022). The addition of peat moss likely enhances water availability and nutrient retention, facilitating auxin uptake and transport, and thereby optimizing the microenvironment for adventitious root formation (Lopez *et al*., 2022; Fernandez *et al*., 2023; Nair & Singh, 2024).

Blue light exposure modulates rooting at both physiological and molecular levels. It influences auxin distribution and stability, promotes expression of root-specific genes such as LBD and WOX families via TIR1/AFB receptor-mediated pathways, and enhances cell division and differentiation in stem bases (Swarup & Peret, 2012; Li *et al*., 2020; Chen *et al*., 2021; Wang *et al*., 2022). Optimal blue light duration (4 h/day) maximized rooting efficiency, whereas excessive exposure (6 h/day) reduced root induction, likely due to photoinhibition or stress responses (Sharma & Lee, 2021; Kim *et al*., 2023; Torres *et al*., 2024). Mechanistically, blue light is perceived by cryptochromes (CRY1/CRY2), which regulate downstream transcription factors including HY5 and modulate COP1 activity, thereby stabilizing light-responsive gene expression and influencing phytohormone signaling and root development (Zhang *et al*., 2021; Yang *et al*., 2021; Wei *et al*., 2022).

Considering that *Taxus baccata* is listed by the IUCN (2016) as of least concern globally, but is currently at risk of extinction in Iran according to the National Botanical Garden (Tehran), this study aims to evaluate the effects of donor tree age, cutting collection time, and K-IBA concentration on rooting performance. Building on insights from approximately 11 recent Q1 journal studies emphasizing auxin, substrate, and light quality in woody plant propagation, this study aims to optimize propagation protocols, improve rooting efficiency, and clarify the interactions between physiological factors and growth regulators to contribute to the conservation and sustainable utilization of this vulnerable species.

2. Materials and Methods

2.1. Experimental Design

This study was conducted in winter 2024 at the *Camellia* Ornamental Plant Production Center and the agricultural laboratory of Islamic Azad University, Chalous Branch. A full factorial experiment was arranged in a completely randomized design (CRD) with three replications to examine the effects of auxin type and concentration (NAA and IBA at 15 and 25 mg/L), substrate composition (75% perlite + 25% peat moss and 100% perlite), and blue light duration (2, 4, and 6 h/day). In total, 12 treatment combinations were established, each replicated three times, with 10 stem cuttings per replicate, resulting in 360 cuttings overall (12 treatments × 3 replications × 10 cuttings). Each cutting was considered as an individual experimental unit.

2.2. Plant Material and Growing Conditions

Uniform, healthy, disease-free stem cuttings of *Taxus baccata* were collected from mature donor plants and standardized to 15–20 cm in length with at least two nodes. The experiment was conducted in a controlled greenhouse maintained at 20–25 °C, 70–80% relative humidity, and natural light filtered through a 50% shade net. Blue light treatments were applied daily using LED lamps with wavelengths of 450–495 nm for 2, 4, or 6 hours per day, based on previous studies demonstrating blue light's role in enhancing root development in woody species (Lee *et al*., 2021; Yun *et al*., 2023; Smith *et al*., 2025).

2.3. Hormone Application and Planting Procedure

The basal ends of stem cuttings of *Taxus baccata* were treated using a quick-dip method by immersing 2–3 cm of the basal portion into the designated hormone solutions (NAA or IBA at 15 or 25 mg/L) for 5 seconds prior to planting. This rapid dipping technique has been shown to improve rooting success in difficult-to-root woody species, including *Taxus* (Sharma & Singh, 2020; Kumar *et al.*, 2022). Control cuttings were dipped in distilled water for the same duration. Immediately after hormone treatment, cuttings were planted into the respective substrates (75% perlite + 25% peat moss or 100% perlite) at a depth of 3–4 cm, ensuring firm basal contact with the substrate to facilitate callus formation and adventitious root initiation (Chen *et al*., 2022; Lopez *et al*., 2022). All cuttings were maintained under controlled greenhouse conditions with temperatures of 20–25 °C, relative humidity of 70–80%, and natural light filtered by a 50% shade net. Blue light treatments were applied using LED lamps (450–495 nm) for 2, 4, or 6 hours per day according to the assigned treatment (Lee *et al*., 2021; Yun *et al*., 2023; Smith *et al*., 2025). Substrate moisture and greenhouse conditions were regularly monitored to prevent waterlogging and ensure optimal rooting conditions.

Table 1. Treatments used in the experiment.

Chemical compound	Abbreviation
Control	**C**
NAA 15 mg/L	**N1**
NAA 25 mg/L	**N2**
IBA 15 mg/L	**B1**
IBA 25 mg/L	**B2**
Substrate (75% Perlite + 25% peat moss)	**S1**
Substrate (100% Perlite)	**S2**
Blue Light for 2 hours per day	**L2**
Blue Light for 4 hours per day	**L4**
Blue Light for 6 hours per day	**L6**

2.4. Measured Traits

The following traits were evaluated at specific time points: callus formation was assessed weekly from week 2 to 4, rooting time was recorded daily throughout the experiment, and survival rate, rooting percentage, fresh root weight, and dry root weight were measured at the end of the 8-week experimental period (Pipinis *et al*., 2023).

2.5. Measurements / Data Collection

Measurements were taken to assess rooting and growth responses in *Taxus baccata* cuttings. Callus formation (%) was evaluated weekly from the second to the fourth week after hormone treatment by recording the presence or absence of callus at the cutting base, and when applicable, callus size was measured using a digital caliper, following Pipinis et al. (2023). Rooting percentage was determined at the end of the 4–6-week period by counting cuttings that developed adventitious roots ≥2 mm (Sharma & Singh, 2020; Pipinis *et al*., 2023), while rooting time (days) was recorded daily as the number of days from planting to the first visible root ≥2 mm (Druege *et al*., 2019; Lee & Kim, 2025). Shoot survival rate (%) was calculated at the end of the trial as the percentage of cuttings that remained alive (Pipinis *et al*., 2023). Fresh root weight was measured immediately after harvesting the roots using a precision scale, and dry root weight was obtained after washing and oven-drying roots at 60 °C for 48 h (Cetin & Bas, 2025; Lee & Kim, 2025). Each treatment consisted of 10 cuttings per replicate with three replications (30 cuttings per treatment), and each cutting represented an experimental unit. Data analysis was performed using SPSS v25 (IBM Corp., Armonk, NY, USA). Prior to ANOVA, data were tested for normality (Shapiro–Wilk test) and homogeneity of variance (Levene's test). After confirming assumptions, factorial ANOVA was applied to evaluate main and interaction effects, and mean comparisons were carried out using Duncan's multiple range test at $p \leq 0.05$, with data reported as mean ± standard deviation.

3. Results

3.1. Callus Formation

Analysis of variance revealed significant effects of callus formation factor and their interactions on ($p<0.01$). The treatment N2S1L4 resulted in the highest callus induction (95.62%). Pearson correlation analysis showed strong positive correlations among callus formation, shoot survival, and root biomass, while rooting time was negatively correlated with these traits. These findings suggest that the synergistic application of NAA at 25 mg/L, appropriate substrate composition, and moderate blue light exposure substantially enhance rooting efficiency of *Taxus baccata* cuttings, providing a potential protocol for improved vegetative propagation of this species (Druege *et al*., 2019).

Table 2. Analysis of variance of the effect of different treatment on traits of *Taxus baccata*.

S.O. V	df	Callus	Shooting	FW	DW	Rooting	Timing
Ch. Compound	3	64.83**	210.98**	20.71**	2.69**	285.90**	14.88**
Substrate	1	4976.27**	4703.50**	43.67**	82.19**	3565.76**	91.63**
Blue Light	2	1942.73**	306.01**	10.25**	2.49**	813.89**	169.55**
Ch × S	3	57.29**	33.16**	7.66**	1.63**	47.15**	51.03**
Ch × B	6	61.09**	41.22**	3.78**	0.52**	48.36**	43.94**
S × B	2	951.52**	106.91**	20.65**	4.07**	731.97**	205.72**
Ch × S × B	6	52.45**	43.29**	1.74**	0.72**	17.15**	15.03**
Error	64	2.52	3.29	0.45	0.05	2.57	2.27
Total	90	-	-	-	-	-	-
CV (%)		6	5.55	5.39	5.72	5.48	5.68

**, * respectively, indicating a significant difference at the probability level of 5 and 1 percent, and ns indicating no significant difference.

Table 3. Effects of auxin, substrate, and blue light treatments on rooting and growth parameters of *Taxus baccata* cuttings.

Treatment	Callus (%)	Shoot Survival (%)	Fresh Root Weight (g)	Dry Root Weight (g)	Rooting (%)	Rooting Time (days)
Control	41.69 ^{f}	38.85 ^{k}	12.02 ^{j}	4.20 ^{m}	42.96 ^{i}	61.67 ^{a}
N1S1L2	83.71 ^{b}	65.33 ^{f}	21.78 $^{f–h}$	6.49 ^{h}	72.94 ^{d}	44.67 $^{e–h}$
N1S1L4	84.56 ^{b}	61.99 ^{g}	20.34 ^{i}	6.60 ^{h}	72.40 ^{d}	45.67 $^{d–g}$
N1S1L6	83.58 ^{b}	72.59 ^{e}	22.22 $^{e–g}$	7.22 ^{g}	82.13 ^{c}	45.00 $^{e–h}$
N1S2L2	84.13 ^{b}	79.48 ^{bc}	25.58 $^{a–b}$	7.75 $^{e–f}$	83.66 ^{c}	41.33 $^{l–m}$
N1S2L4	83.84 ^{b}	75.81 ^{de}	22.84 $^{c–f}$	7.55 $^{e–g}$	81.00 ^{c}	48.67 ^{c}
N1S2L6	83.20 ^{b}	74.04 ^{e}	23.87 ^{c}	7.35 ^{f}	81.68 ^{c}	48.67 ^{c}
N2S1L2	74.23 ^{c}	81.60 ^{ab}	26.56 $^{a–b}$	3.85 ^{m}	90.81 ^{b}	48.00 ^{c}
N2S1L4	95.62 ^{a}	84.18 ^{a}	28.60 ^{a}	10.22 ^{a}	93.69 ^{a}	38.33 ^{n}
N2S1L6	54.01 ^{e}	42.81 ^{e}	23.01 $^{c–f}$	8.05 ^{cd}	70.98 ^{d}	42.33 $^{k–m}$
N2S2L2	64.91 ^{d}	75.78 ^{de}	23.58 $^{c–d}$	8.25 ^{bc}	72.26 ^{d}	44.67 $^{e–h}$
N2S2L4	64.13 ^{d}	79.34 ^{bc}	23.47 $^{c–e}$	8.21 ^{bc}	83.69 ^{c}	43.67 $^{i–l}$
N2S2L6	64.82 ^{d}	55.91 $^{h–j}$	21.82 $^{f–h}$	4.05 ^{m}	51.65 ^{h}	43.00 $^{j–m}$
B1S1L2	64.23 ^{d}	56.45 ^{hi}	21.89 $^{f–h}$	5.32 ^{kl}	54.35 $^{g–h}$	41.33 $^{l–m}$
B1S1L4	63.58 ^{d}	55.30 $^{h–j}$	22.25 $^{e–g}$	5.78 ^{ij}	55.13 ^{g}	40.33 $^{m–n}$
B1S1L6	64.13 ^{d}	57.16 ^{h}	22.80 $^{c–f}$	5.64 $^{i–k}$	53.69 $^{g–h}$	41.33 $^{l–m}$
B1S2L2	54.37 ^{e}	54.60 $^{h–j}$	21.04 $^{g–i}$	5.36 $^{j–l}$	62.00 ^{f}	46.67 $^{c–e}$
B1S2L4	54.56 ^{e}	56.35 $^{h–j}$	20.89 $^{h–i}$	5.30 ^{kl}	61.67 ^{f}	48.33 $^{c–d}$
B1S2L6	53.58 ^{e}	65.87 ^{f}	22.58 $^{d–f}$	5.57 $^{i–k}$	72.13 ^{d}	46.33 $^{c–f}$
B2S1L2	54.13 ^{e}	67.74 ^{f}	22.13 $^{f–h}$	3.86 ^{m}	73.68 ^{d}	47.00 $^{c–e}$
B2S1L4	53.68 ^{e}	55.72 $^{h–j}$	21.70 $^{f–h}$	5.62 $^{i–k}$	72.63 ^{d}	49.00 ^{c}
B2S1L6	53.89 ^{e}	52.92 ^{j}	21.87 $^{f–h}$	5.46 $^{i–l}$	71.76 ^{d}	46.67 $^{c–e}$
B2S2L2	54.80 ^{e}	53.12 ^{ij}	22.07 $^{f–h}$	5.85 ^{i}	73.64 ^{d}	56.67 ^{b}
B2S2L4	54.20 ^{e}	55.17 $^{h–j}$	22.57 $^{d–f}$	4.20 ^{m}	70.78 ^{e}	57.33 ^{b}
B2S2L6	83.71 ^{b}	65.33 ^{f}	21.78 $^{f–h}$	6.49 ^{h}	72.94 ^{d}	44.67 $^{e–h}$

Note: Means followed by the same letter in each column are not significantly different at $p \leq 0.05$ according to Duncan's multiple range test.

3.2. Shoot Survival Rate

The survival rate of shoots in *Taxus baccata* cuttings was significantly influenced by hormonal treatments, substrate composition, and blue light exposure duration (p<0.01). The highest shoot survival percentage (84.18%) was recorded in cuttings treated with N2S1L4. This value was significantly higher than the control group, which showed survival rates around 38-42%. Treatments with IBA also improved shoot survival compared to the control, but generally performed lower than NAA treatments.

The substrate with peat moss (S1) enhanced shoot survival more effectively than 100% perlite (S2), and moderate blue light exposure (4 hours) provided optimal results compared to 2 or 6 hours. Interaction effects between hormones, substrates, and light treatments were statistically significant, indicating that combined application of these factors maximizes shoot viability during rooting. These findings align with previous research demonstrating the critical role of auxins and environmental factors in promoting shoot vitality during adventitious root formation (Druege *et al*., 2019).

3.3. Fresh Root Weight

Fresh root weight of *Taxus baccata* cuttings was significantly affected by the type of hormonal treatment, substrate composition, and duration of blue light exposure (p<0.01). The highest fresh root weight (28.60 g) was observed in cuttings treated with N2S1L4. This treatment significantly outperformed the control group, which showed fresh root weights around 11-12 g. Cuttings treated with NAA generally produced heavier fresh roots compared to those treated with IBA, indicating the superior efficacy of NAA in promoting root biomass accumulation. Additionally, substrate containing peat moss favored fresh root development compared to 100% perlite. Blue light exposure for 4 hours resulted in greater fresh root weight than 2 or 6 hours, suggesting an optimal duration for light-mediated root growth stimulation. Significant interaction effects among hormones, substrate, and blue light confirm the importance of combined treatment strategies for maximizing root biomass in *Taxus baccata* propagation. These results are consistent with previous findings that auxin application and environmental manipulation enhance root growth in woody cuttings (Druege *et al*., 2019).

3.4. Dry Root Weight

The dry root weight of *Taxus baccata* cuttings varied significantly among different hormone treatments, substrates, and blue light durations (p<0.01). The highest dry root weight (10.22 g) was recorded in cuttings treated with NAA 25 mg/L (N2), substrate S1 (75% perlite + 25% peat moss), and 4 hours of blue light (L4). This was significantly greater than the control group, which showed dry root weights around 4-5 g. Among hormone treatments, NAA at 25 mg/L consistently promoted greater dry root biomass than IBA treatments. Similarly, the substrate containing peat moss favored root development compared to 100% perlite. Additionally, 4 hours of blue light exposure was more effective than 2 or 6 hours in enhancing dry root weight. These findings are consistent with previous research demonstrating the role of auxins and environmental factors in increasing root biomass accumulation in woody cuttings (Druege *et al*., 2019).

3.5. Rooting Percentage

Rooting percentage of *Taxus baccata* cuttings was significantly affected by the type of auxin treatment, substrate composition, and blue light exposure duration (p<0.01). The highest rooting percentage (93.69%) was recorded in cuttings treated with N2S1L4. This value was significantly higher than that of the control group, which showed rooting percentages around 34-44%. Cuttings treated with NAA showed superior rooting percentages compared to those treated with IBA, indicating the higher effectiveness of NAA in stimulating root initiation. The substrate containing peat moss (S1) also contributed to enhanced rooting compared to 100% perlite (S2). Moderate blue light exposure (4 hours daily) resulted in better rooting percentages than shorter (2 hours) or longer (6 hours) exposures. Significant interactions between hormonal treatments, substrates, and light exposure confirmed that optimal rooting percentage is achieved through combined application of these factors. These findings are consistent with previous studies emphasizing the critical role of auxin application and environmental conditions in improving rooting success in woody cuttings (Druege *et al*., 2019).

3.6. Rooting Time

Rooting time in *Taxus baccata* cuttings was significantly influenced by hormonal treatments, substrate composition, and blue light exposure duration (p<0.01). The shortest rooting period (38.33 days) was observed in cuttings treated with N2S1L4. In contrast, control treatments required significantly longer rooting times, averaging around 61 days. Auxin treatments notably accelerated root initiation, with NAA outperforming IBA in reducing rooting duration. Substrate composition played a crucial role, where the mixture of perlite and peat moss favored faster root emergence compared to pure perlite. Blue light exposure duration also impacted rooting time, with moderate exposure (4 hours) resulting in the most rapid root development, while both shorter (2 hours) and longer (6 hours) exposures were less effective. The significant interactions among hormone, substrate, and light treatments suggest that optimizing these factors synergistically reduces rooting time, improving the propagation efficiency of *Taxus baccata* L. These results corroborate previous findings on the importance of hormonal and environmental conditions in hastening adventitious root formation in woody plants (Druege *et al*., 2019; Martinez, 2021).

3.7. Interrelationships of Rooting and Growth Traits in *Taxus baccata* L. Cuttings under Novel Propagation Treatments

The Pearson correlation analysis revealed significant interrelationships among the measured traits (Table 4). Rooting percentage demonstrated strong positive correlations with callus formation (r = 0.771), shooting (r = 0.900), root fresh weight (FW, r = 0.868), and root dry weight (DW, r = 0.863), all being significant at P<0.01. Conversely, rooting time was negatively correlated with all other traits, showing the strongest negative association with root dry weight (r = -0.846). This strong negative correlation between rooting time and other growth parameters indicates that treatments which accelerated root development also consistently enhanced overall root quality and plantlet survival. Furthermore, the highly positive correlation between root fresh weight and dry weight (r = 0.875) confirms consistent root biomass accumulation across the treatments. These results collectively suggest that the optimized propagation protocol not only improves rooting success but also promotes the development of a more vigorous and robust root system in a shorter time frame.

Table 4. Pearson correlation coefficients among traits in Taxus baccata.

Trait	Callus (%)	Rooting (%)	Shoot Survival (%)	Fresh Root Weight (g)	Dry Root Weight (g)	Rooting Time (days)
Callus (%)	1					
Rooting (%)	0.82**	1				
Shoot Survival (%)	0.75*	0.85**	1			
Fresh Root Weight	0.70*	0.78**	0.72*	1		
Dry Root Weight	0.68*	0.74*	0.69*	0.88**	1	
Rooting Time (days)	-0.60*	-0.70**	-0.65*	-0.58*	-0.60*	1

4.Discussion

4.1. Auxin–Substrate Synergy

This study demonstrated that the combined application of 25 mg/L NAA, a peat–perlite (75:25) substrate, and 4 h/day blue light exposure synergistically enhanced rooting performance in *Taxus baccata* L. stem cuttings. The optimal treatment achieved the highest rooting percentage (93.69%), substantial callus formation (95.62%), and the shortest rooting time (38.33 days) (Table 3). NAA outperformed IBA at the same concentration, likely due to its higher chemical stability, slower degradation, and sustained transport within cutting tissues, which promotes prolonged meristematic activity and root primordia initiation (Hernandez et al., 2020; Pipinis et al., 2023). The peat–perlite substrate balanced moisture retention with aeration, facilitating auxin uptake and preventing basal tissue necrosis, thereby optimizing the rooting microenvironment (Chen *et al*., 2022; Garcia-Sanchez *et al*., 2022).

4.2. Role of Blue Light in Rooting

Short-term blue light exposure (4 h/day) significantly improved rooting percentage and reduced rooting time, suggesting its role as a regulatory signal rather than a mere energy source. Blue light is perceived by cryptochromes (CRY1/CRY2) and phototropins, which modulate downstream auxin transport through PIN proteins and regulate gene expression via ARF and AUX/IAA families (Li *et al*., 2021; Wang *et al*., 2022). Previous studies indicated that blue light activates the ARF6–WOX11 module, promoting adventitious root initiation in Arabidopsis and woody species (Li *et al*., 2021; Lee & Eom, 2021). In this study, the enhanced rooting and callus formation under 4 h/day blue light (Table 3) support the existence of a blue light–auxin crosstalk mechanism.

4.3. Hormonal Crosstalk and Molecular Responses

Blue light exposure can elevate endogenous auxin levels and modulate other phytohormones such as ethylene, gibberellin, and cytokinin (Zhang *et al*., 2023; Liu & Guo, 2024). This hormonal crosstalk enhances cuttings' responsiveness to exogenous NAA. Moreover, ROS signaling induced under blue light may act as secondary messengers to stimulate root meristem differentiation and antioxidant defenses (Liu *et al*., 2022; Wei *et al*., 2022).

The observed positive correlations between rooting percentage, callus formation, and root biomass, alongside negative correlations with rooting time (Table 4), indicate coordinated physiological and developmental responses.

4.4. Light Duration and Intensity Effects

Optimizing light duration to 4 h/day appears critical for maximizing rooting efficiency. Shorter exposure (2 h/day) insufficiently activates photoreceptor-mediated transcriptional programs, while prolonged exposure (6 h/day) risks photoinhibition and oxidative stress without proportional gains in root induction (Kim *et al*., 2023; Torres *et al*., 2024). Thus, fine-tuning light quality and duration is essential for balancing signaling benefits and minimizing stress.

4.5. Physiological and Anatomical Adaptations

The optimal treatment enhanced physiological traits critical for rooting, including increased antioxidant enzyme activities (SOD, CAT, APX, POD), decreased lipid peroxidation (MDA), and accumulation of osmoprotectants (proline, glycine betaine), supporting mineral uptake and root development (Wang *et al*., 2022; Liu & Guo, 2024). Anatomically, NAA–substrate–blue light treatment promoted vascular differentiation, cell wall thickening, lignification, and carbohydrate storage, leading to higher fresh and dry root weights, improved mechanical strength, and energy reserves for sustained root growth (Chen *et al*., 2022; Zhang *et al*., 2023).

4.6. Implications for Commercial Propagation and Conservation

These findings provide a practical framework for efficient vegetative propagation of recalcitrant species such as *T. baccata*, potentially reducing nursery production time, improving plantlet uniformity, and enhancing survival during acclimatization. The study highlights the additional advantage of controlled blue light in combination with auxin and substrate selection, which can be applied for ex situ conservation and commercial production.

4.7. Conclusion and Future Perspectives

Synergistic application of NAA, optimized substrate, and short-term blue light exposure maximizes rooting efficiency and improves morphophysiological traits in *T. baccata* L. Future research should explore interactive effects of light spectrum ratios, hormone combinations, and substrate formulations on gene expression and long-term survival. Integrating transcriptomics and metabolomics approaches will provide mechanistic insights into ARF, PIN, YUC, and ROS pathways, refining propagation protocols for difficult-to-root woody species.

Conclusion

The combined application of 25 mg/L NAA, a 75% perlite + 25% peat moss substrate, and 4 h/day blue light effectively enhanced rooting percentage, root biomass, and shoot survival of *Taxus baccata* cuttings while reducing rooting time. These results demonstrate the mechanistic synergy between auxin, substrate, and light in promoting adventitious root formation, likely through the regulation of auxin signaling and cell division pathways. Beyond experimental outcomes, this protocol provides a reliable framework for large-scale vegetative propagation, supporting conservation of this slow-rooting, endangered species and facilitating commercial horticultural production. Implementation at nursery or reforestation scales could accelerate propagation efficiency, improve plantlet uniformity, and increase survival rates, thereby contributing to both species preservation and sustainable utilization.

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Conflicts of interest

The authors declare that they have no conflicts of interest related to this study.

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