Journal of Applied Botany and Food Quality 98, 147 - 157 (2025), DOI:10.5073/JABFQ.2025.098.017

[1]Research Center for Applied Botany, National Research and Innovation Agency, Bogor, West Java, Indonesia.
[2]Research Center for Nuclear Beam Analysis Technology, National Research and Innovation Agency, Bandung, West Java, Indonesia
[3]Research Center for Photonics, National Research and Innovation Agency, South Tangerang, Indonesia
[4]Research Center for Applied Microbiology, National Research and Innovation Agency, Bogor, West Java, Indonesia
[5]Research Center for Biomass and Bioproducts, National Research and Innovation Agency, Bogor, West Java, Indonesia
[6]Research Center for Biota Systems, National Research and Innovation Agency, Bogor, West Java, Indonesia
[7]Departement of Biology, Faculty of Mathematics and Natural Sciences, IPB University, Bogor, West Java, Indonesia
[8]Biotech Center, Collaborative Research Center, IPB University, Dramaga Bogor, Indonesia

Artificial light at night (ALAN) induces different photosynthetic activities and secondary metabolite accumulation in several plant species

Mutiara K. Pitaloka[1*], Kartika Ning Tyas[1], Muhayatun Santoso[2], Isnaeni Isnaeni[3], Andes Hamuraby Rozak[1], Ahmad Fathoni[4], Sukma Surya Kusumah[5], Rizmoon N. Zulkarnaen[1], Didi Usmadi[6], Dwi Setyo Rini[1], Triadiati Triadiati[7,8], Maya Ismayati[5], Frisca Damayanti[1], Reza Ramdan Rivai[1]

(Submitted: March 1, 2024; Accepted: August 31, 2025)

Summary

It is well established that nighttime light exposure can negatively influence the physiological performance of plants. Nevertheless, the specific effects of artificial light at night (ALAN) on plants under natural conditions remain insufficiently understood. The Bogor Botanic Gardens (BBG) is widely recognized as an ex-situ conservation site for diverse plant species. Although BBG provides an ecosystem resembling natural habitats, its location within the center of a rapidly developing urban area exposes plant collections to direct and indirect anthropogenic pressures, including light pollution. In this study, we examined the impact of ALAN installations at BBG on fourteen plant species by quantifying several physiological parameters, including photosynthetic activity, stomatal conductance, chlorophyll content, secondary metabolite accumulation, and mineral nutrient concentrations. Our results demonstrated significant alterations in photosynthesis (A) and stomatal conductance (g_{sw}) in several species exposed to ALAN. In particular, we focused on one secondary metabolite, neophytadiene, and observed that its content was consistently higher after treatments (T1 and T2) compared to pre-treatment levels (T0) in most of the tested species, indicating a measurable effect of ALAN. Furthermore, we detected a pronounced reduction in iron (Fe) concentration across five plant species, although this decrease did not appear to be directly associated with changes in photosynthetic performance. These findings provide valuable insights into the physiological responses of plants to ALAN and may inform strategies for plant selection in urban environments. Such strategies could help alleviate the adverse effects of light pollution on plant physiological functions and improve the sustainability of urban green spaces.

Keywords: ALAN, Photosynthesis, Plant mineral nutrients, Stomata conductance, Secondary metabolites

Inroduction

Light is a key factor influencing plant growth and development, as plants use complex pigment systems and photoreceptors to absorb specific wavelengths of the spectrum, triggering photosynthesis as well as photomorphogenic and phototropic responses that regulate their physiological and metabolic functions(Farquhar and Sharkey, 1982; Fukuda, 2013; Weller and Kendrick, 2008). In higher plants, photosynthetically active radiation (PAR) which is a portion of sunlight with wavelengths between 400 and 700 nm, is collected by photosynthetic pigments in the photosystem PSI and PSII (Wimalasekera, 2019). These light-dependent processes not only fuel energy capture but also synchronize plants with the natural day and night cycle, ensuring that physiological functions align with environmental cues.

Exposure to ALAN during the night can disrupt the circadian rhythm of plants, either by slowing down or speeding up their cycles. As a result, ALAN may act as a false signal, creating the impression of extended daylight for plants (Basler and Körner, 2014). Circadian rhythm is a diurnal time response of plants in which the cycles last approximately 24 hours and are synchronized with light and dark cycles of the rotating planet as well as response to abiotic and biotic factors (Venkat and Muneer, 2022). Such alterations to natural night conditions are expected to influence the physiological functions of many organisms, including plants. It has been suggested that the dark period is crucial for plants because their physiological responses to the environment depend on both light and darkness (Gaston et al., 2013). Moreover, it has been suggested that trees and shrubs grown under streetlights exhibit longer growing seasons, earlier leaf emergence, and later leaf fall than those that stayed in darker environments (Singhal et al., 2019). Even though the intensity of some ALAN such as those from streetlights or vehicle headlights may be considered low, it still can induce physiological effects in plants and sustained exposure at these levels may potentially contribute to substantial ecological disruptions (Bennie et al., 2016; Poulin et al., 2014). Previous study also show that ALAN significantly disrupts photosynthesis in urban plants *Euonymus japonicus* and *Rosa hybrida* by reducing photosystem II efficiency, electron transport, photochemical quenching, and net photosynthetic rate, while also lowering stomatal conductance and gas exchange capacity (Wei et al., 2023). Night-time light perceived by phytochromes can disrupt the transmission of environmental information, generating misleading signals that interfere with plant signaling networks and physiological regulation, thereby compromising their functional performance during the day(Gaston et al., 2013).

It has been widely understood that ALAN might harm the natural organism negatively (Dalle Carbonare et al., 2023; Singhal et al., 2019). Despite this, only a limited number of researchers have at-

* Corresponding author

tempted to investigate ALAN in plants and assess its physiological features in natural settings. Bogor Botanic Gardens (BBG), founded in 1817, is the oldest botanical garden in Southeast Asia and holds a significant place in the country's history(ARIATI and WIDYATMOKO, 2019). BBG plays a crucial role in plant conservation, research activities, education programs, ecotourism opportunities, and environmental services. Additionally, some ALAN installations were also found in several areas within BBG either for tourism attraction or road lighting purposes, which caused concern among many. Click or tap here to enter text.

Given these concerns, it becomes essential to understand how prolonged exposure to ALAN influences plant physiology. Prolonged illumination can disrupt natural light-dark cycles, altering physiological performance and inducing abiotic stress that leads to the accumulation of reactive oxygen species (ROS) and changes in secondary metabolite production (SANKHUAN et al., 2022; ZHANG et al., 2021). Light intensity, spectrum, and photoperiod are known to shape secondary metabolite accumulation, high light enhances flavonoid production in *Lonicera japonica* (FANG et al., 2020), while specific light qualities regulate terpenoid biosynthesis in *Artemisia annua* (SANKHUAN et al., 2022). Therefore, in this study we aim to evaluate the effects of ALAN on photosynthesis, physiological traits, and the accumulation of secondary metabolites and mineral nutrients in plant collections at Bogor Botanic Gardens. To achieve this, we selected fourteen plant species representing different growth forms exposed to ALAN and measured their physiological properties as model systems.

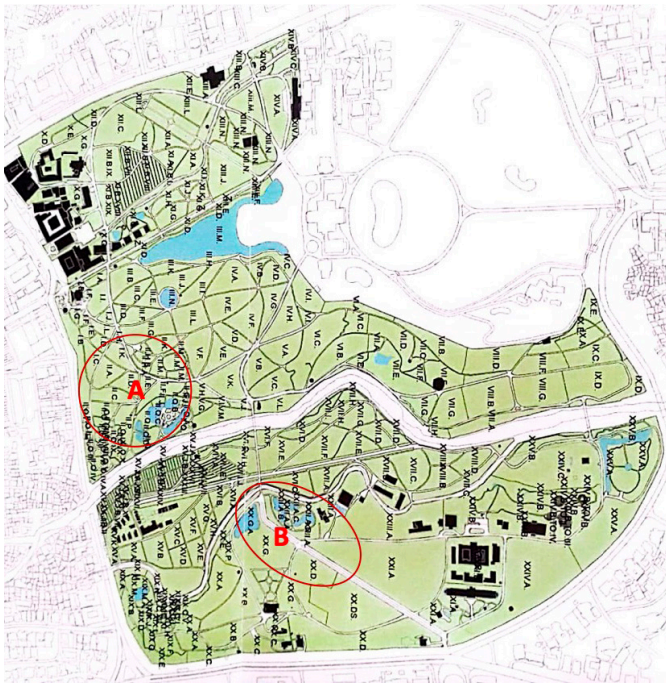

Fig. 1: Map of BBG and location of ALAN installation (Red circle) within BBG.

Material and methodology

ALAN installation and sample selection

Several types of LED lights were installed in the BBG area, along with plant collections located in sections X.G to XXI.A of the BBG. The types of lights and their installation sites are summarized in Tab. S1. Fourteen plant species were randomly selected to represent different growth forms (habitus). For each species, one treatment plant (exposed to artificial light at night, ALAN) and one control plant (not exposed to ALAN) were measured. Because ALAN installations in the BBG were limited to specific species and areas, all species had no replicated treatments. The lights were switched on twice a week for up to 4 hours per day, from 6:00 p.m. to 11:00 p.m. Random measurements of LED light intensity within the BBG area showed that most plants were exposed to less than 20 lumens. *Barringtonia asiatica, Dalbergia cultrata, Dillenia indica, Diospyros blancoi Hanguana malayana, Inocarpus fagifer, Pandanus tectoris, Parmentiera aculeata, Phoenix reclinate, Posoquera latifolia, Salacca salacca, Syzygium semarangense* located at region A (Fig. 1). *Agathis damara* and *Canarium indicum* located at region B. Individual plants are depicted in Fig. 2.

Experimental set up of ALAN to *Dyospiros blancoi*

Seedlings of *Diospyros blancoi* were used in the experiment. An enclosed growth chamber measuring 2 × 2 m was constructed from light steel. LED lamps emitting flashing colors (red, green, and blue) were installed at the top of the chamber. Seedlings were placed on shelves inside, and the lamps were programmed to switch on daily at 18:00 with two light duration; 1 and 12 hours. The other setup with 6 hours light where light was witch on in every two and seven days. Some Control plants were grown in an identical chamber without artificial light installation. These plants were exposed only to natural light conditions, experiencing daylight during the day and complete darkness at night. After four months of exposure to artificial light at night (ALAN), gas exchange measurements were conducted on the leaves to assess the photosynthetic activity of the seedlings.

Leaf gas exchange analysis and chlorophyll measurement

Leaf gas exchange was measured twice, the first measurement (T_1) was done in May 2023- 4 months after ALAN was first started. While second measurement (T_2) was done in September 2023- 8 months after ALAN started. Infra-red gas (IRGA) analysis was conducted using a Li-Cor 6800 (LI-COR BioScience). Steady-state measurement was done with the chamber flow rate set to 400 μmol s^{-1}, leaf temperature to 32 °C, reference [CO_2] to 400 ppm, light intensity to 2000 μmol m^{-2} s^{-1} PAR and relative humidity 60%. For samples which showed a significant difference in Photosynthesis between T1 and T2 data collection. Similar environmental set up was applied to leaf gas exchange measurement at experimental plants (*D. blancoi* seedling) accordingly.

Secondary metabolite analysis

Secondary metabolite analysis was done in three time periods, the first sample collection was collected before the ALAN schedule was started, the second analysis was done in four months and the last analysis was done eight months after ALAN was first turned on. We collected approximately 100-150 grams of fresh weight of leaves sample. For sample preparation, the leaves were washed to clean from any dust and dirt. The leaves were dried in the oven at 60 °C until they reached a constant weight and ground to produce powdered simplicial. Approximately 5 g of the powder then soaked in 50 ml of 96% ethanol for 24 hours and filtered using filter paper. The sample then was evaporated till dry and the extracts were analyzed by gas chromatography-mass spectrometry (GC-MS) (GCMS-QP 2010S, Shimadzu, Kyoto, Japan) using a nonpolar column in the form of Rtx 5. For splitless injection, 5 μL of the extract was injected into an injection port liner at 300 °C. The column was held at 70 °C for 5 minutes and then increased to 230 °C. Helium gas was used as the mobile phase. The detector temperature used was 300 °C, with the MS detector electron energy setting around 70 eV. The libraries used were WILEY229 and NIST62.

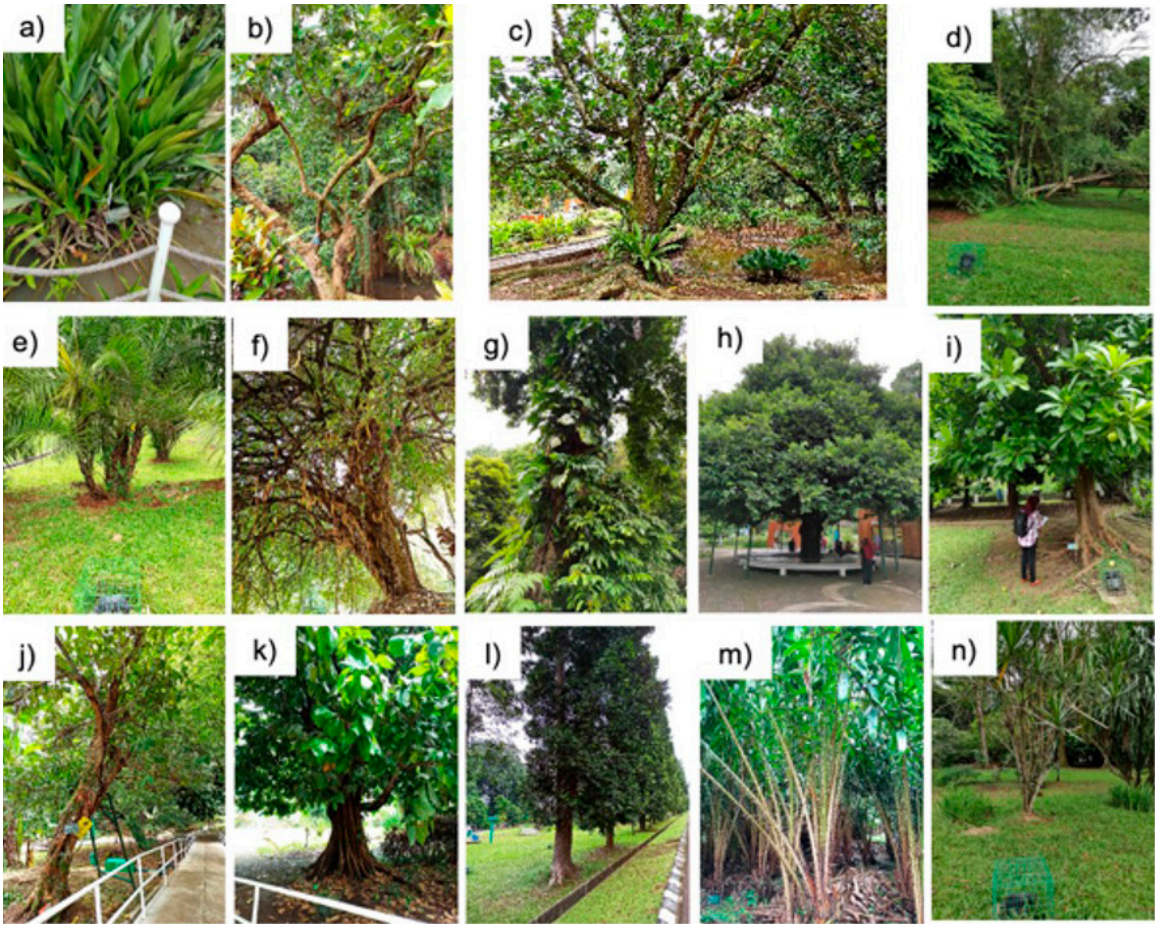

Fig. 2: Plant exposed to ALAN at Bogor Botanic Gardens, (a) *Agathis damara*, (b) *Barringtonia asiatica*, (c) *Canarium indicum*, (d) *Dalbergia cultrata,* (e) *Dillenia indica*, (f) *Diospyros blancoi*, (g) *Hanguana malayana*, (h) *Inocarpus fagifer*, (i) *Pandanus tectoris*, (j) *Parmentiera aculeata*, (k) Phoenix reclinate, (l) *Posoquera latifolia*, (m) *Salacca salacca*, (n) *Syzygium semarangense*.

Plant mineral nutrients analyses

The samples were immediately transported to the laboratory, where upon arrival they were rapidly frozen in liquid nitrogen to halt metabolic activity. They were stored under frozen conditions until further processing. The leaf samples were then crushed using a titanium blade blender (SB TI1726, Sharp Corporation, Sakai, Japan). The mashed sample was then re-weighed and frozen in a freezer (Electrolux, Stockholm, Sweden) at -18 °C. The frozen samples were then dried using a freeze dryer (Alpha 2-4 Martin Christ, Osterode, Germany) at a temperature of -85 °C under vacuum for 48 hours. Dried samples were crushed into fine powders using an agate pestle and mortar, and then placed in a Nalgene bottle. Preparation for a naphthalene acetic acid (NAA) was carried out by weighing 0.05 g of dry, powdered samples using an analytical balance (AG 245 Mettler Toledo Ltd., Melbourne, Australia), which were placed in a 0.3 mL polyethene vial and then sealed by heating. The standard for NAA was also prepared by pipetting 100 µL of ICP multi-element standard solution IV (Merck, Darmstadt, Germany) into a polyethene vial (Cole Palmer, Illinois, USA). The standard in the vial was then dried by an infrared lamp and then sealed by heating (Basuki et al., 2013).

Graph and statistical snalysis

For IRGA measurement data, statistical analysis was conducted using JASP V0.18 (University of Amsterdam, Netherlands). A normality test was done before any statistical analysis was conducted. Classical *t*-tests were performed to determine if there were significant differences between the first and second data collection for a given parameter measured. Graphs were plotted using phyton.

Results

Physiological activity changes on plants after ALAN exposure

Photosynthetic rates (*A*) varied across the 14 species after exposure to ALAN (Fig. 3). Among them, *Dalbergia cultrata* exhibited a significant reduction in *A* at T2 compared to T1, indicating sensitivity to prolonged nighttime light. In contrast, *Diospyros blancoi, Pandanus tectorius,* and *Syzygium samarangense* showed significant increases in *A* at T2, suggesting a potential enhancing effect of ALAN, possibly by extending light availability for carbon fixation. The remaining species, including *Agathis dammara, Barringtonia asiatica, Canarium indicum, Dillenia indica, Hanguana malayana, Inocarpus fagifer, Parmentiera aculeata, Phoenix reclinata, Posoqueria latifolia,* and *Salacca zalacca,* did not exhibit statistically significant differences between treatments or implying relative tolerance or insensitivity to ALAN within the timeframe of observation. Control plants of all species showed no major differences across time points, reinforcing that the observed effects were associated with ALAN rather than other environmental variation.

The effects of ALAN on g_s differed considerably among the studied plants (Fig. 4). In *Dalbergia cultrata*, g_s was significantly lower in treatment plants compared to controls at T1, indicating that ALAN reduced stomatal opening early in the experiment. Similar reductions were observed in *Hanguana malayana* and *Inocarpus fagifer* at T2,

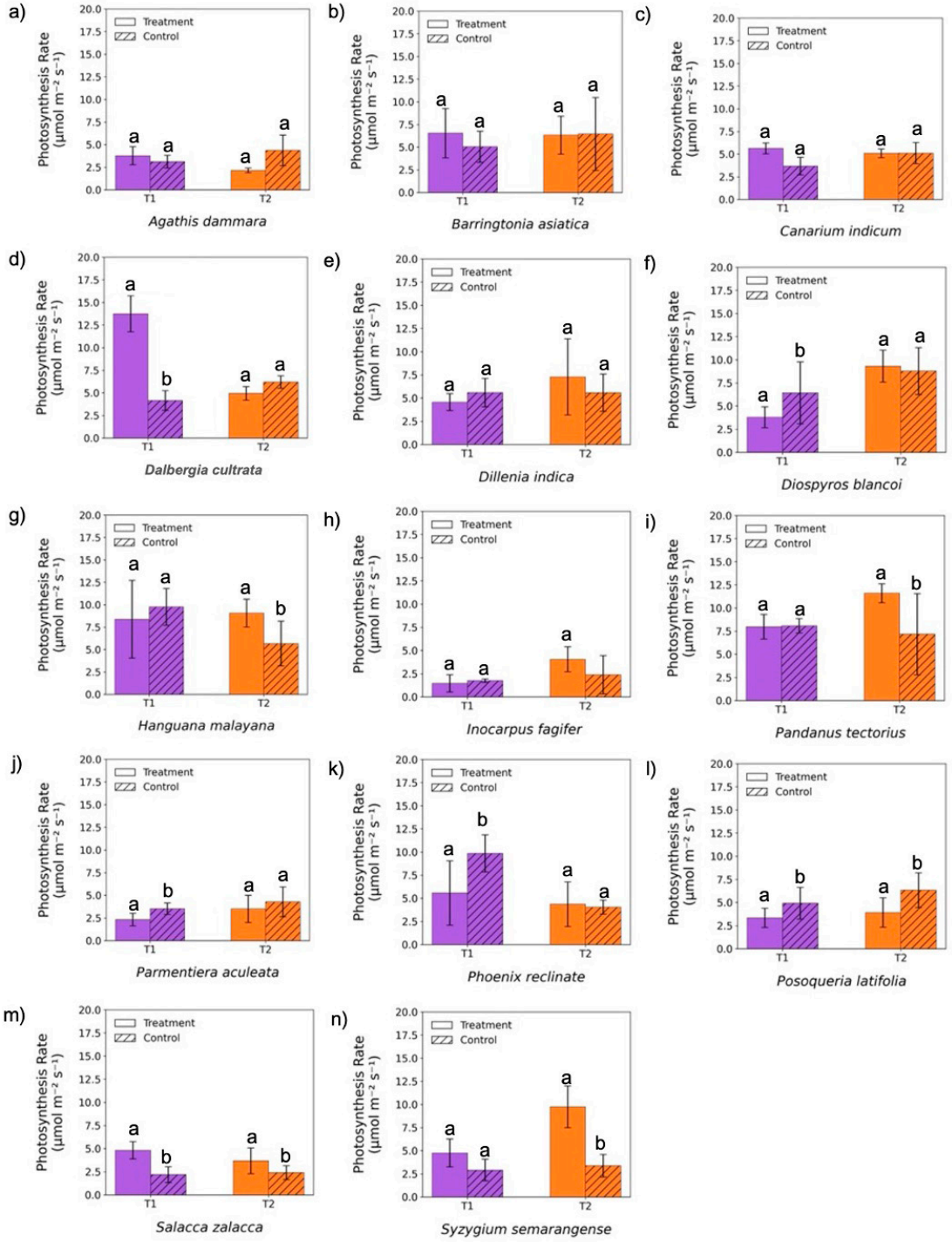

Fig. 3: Photosynthetic rate (A, μmol m^{-2} s^{-1}) of 14 plant species under artificial light at night (treatment) and natural light (control) at two time points (T1 and T2). Bars represent SD. Different letters above bars indicate significant differences between treatments and time points within each species ($p < 0.05$).

suggesting that prolonged exposure to nighttime light constrained g_s in these plants. In contrast, *Syzygium samarangense* exhibited a significant increase in g_s under treatment conditions at T2, indicating that ALAN can also enhance stomatal activity in certain species. Meanwhile other plants, including *Agathis dammara, Barringtonia asiatica, Canarium indicum, Dillenia indica, Parmentiera aculeata, Phoenix reclinata*, and *Salacca zalacca*, showed no significant changes across treatments, demonstrating a relative stability of g_s under both natural and ALAN conditions. Interestingly, *Diospyros blancoi, Pandanus tectorius, and Posoqueria latifolia* displayed an increase in g_s over time in the control plants, but this increase was absent under ALAN, suggesting that ALAN may suppressed g_s. Taken together, these results highlight that while g_s remained largely unaffected in many plants, ALAN exerted both restrictive and enhancing effects depending on species.

Transpiration rate (*E*) increased under treatment in several species, including *Dalbergia cultrata, Diospyros blancoi,* and *Salacca zalacca*, suggesting higher water loss, while in other species such as *Pandanus tectorius* and *Parmentiera aculeata E* remained stable or decreased. Intercellular CO_2 concentration (*Ci*) was higher in treatment plants of *Barringtonia asiatica, Inocarpus fagifer,* and *Syzygium samarangense,* while in *Diospyros blancoi* and *Pandanus tectorius*

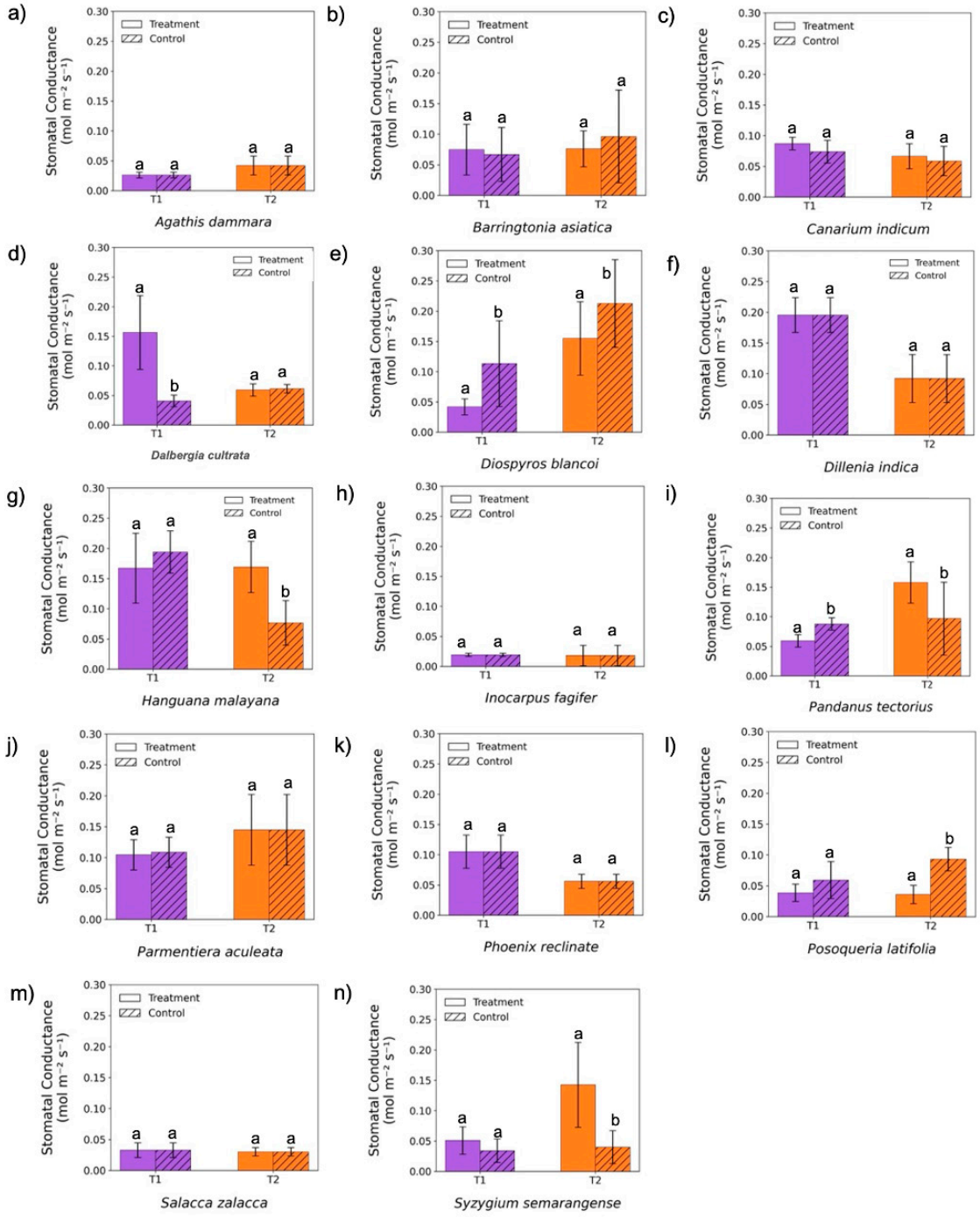

Fig. 4: Stomatal conductance (gs, mol m^{-2} s^{-1}) of 14 plant species under artificial light at night (treatment) and natural light (control) at two time points (T1 and T2). Bars represent SD. Different letters above bars indicate significant differences between treatments and/or time points within each species ($p < 0.05$).

Ci was lower at T2, consistent with greater carbon uptake. Vapor pressure deficit (VPDleaf) and leaf temperature (Tleaf) were mostly stable between control and treatment groups, indicating that ALAN did not substantially affect leaf temperature (Tab. S2).

Additional gas exchange measurements supported these findings (Fig. 5). In treatment plants, *A* and *gs* were significantly higher on Day 2 compared to Day 7 (Fig. 5a-b), indicating an initial ALAN-induced stimulation of photosynthetic performance and stomatal opening that weakened over time. In contrast, control plants showed no significant differences in either *A* (Fig. 5c) or *gs* (Fig. 5d), confirming that gas exchange remained stable under natural conditions. Collectively, these results demonstrate that ALAN can transiently enhance leaf gas exchange in some plants, but this effect is not sustained and varies across species, whereas control plants maintained consistent physiological performance.

ALAN-exposed plants show different secondary metabolite types

In order to better understand the broader stress responses of plants to ALAN, we examined secondary metabolite profiles, as such compounds often exhibit altered accumulation when plants experience environmental stress. While most of the secondary metabolite were recorded in every data collection period (T0, T1, T2), some of them show consistent trends either decrease, increase, or just appear or may

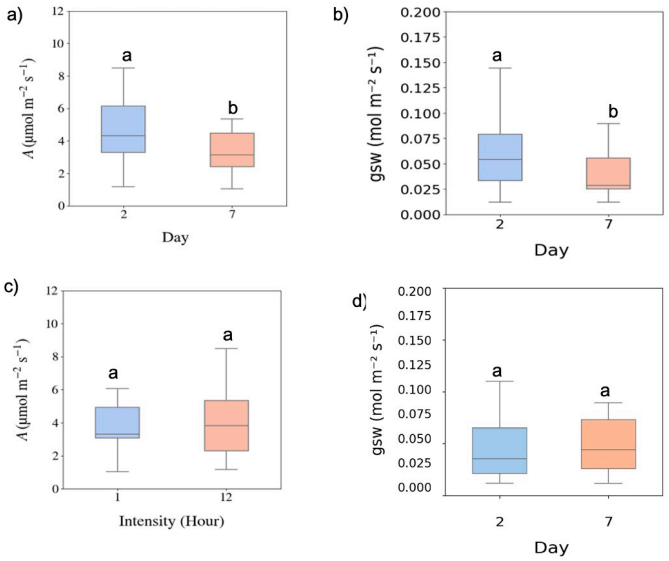

Fig. 5: *A* (μmol m^{-2} s^{-1}) and g_s (mol m^{-2} s^{-1}) of plants under ALAN. (a), *A* in treatment plants measured at Day 2 and Day 7, (b) g_s in treatment plants measured at Day 2 and Day 7, (c) *A* in control plants measured under 1 h and 12 h intensity, (d) g_s in control plants measured at Day 2 and Day 7. Boxplots represent median, interquartile range, and minimum–maximum values. Different letters indicate significant differences between groups ($p < 0.05$).

be completely gone. Secondary metabolite analysis revealed shifts in compound composition under ALAN, with responses varying across plant species (Fig. 6). Several secondary metabolites, including phytol, neophytadiene, squalene, lupeol, and γ-sitosterol, were commonly detected across species. In many plants, levels of these compounds increased at T1 or T2 compared to baseline, suggesting that ALAN promoted secondary metabolite accumulation. For example, in *Agathis damara, Barringtonia asiatica, Dillenia indica, Dalbergia cultrata*, and *Parmentiera aculeata,* higher relative peak areas were observed for sterols and terpenoids under ALAN exposure.

Some species displayed stronger responses than others. *Diospyros blancoi* showed increased levels of catechol, tocopherols, and sterols at later time points, while *Syzygium samarangense* accumulated higher amounts of β-sitosterol, γ-sitosterol, and tocopherols. Similarly, *Hanguana malayana* exhibited increases in phytol, neophytadiene, and tocopherols. By contrast, in species such as *Pandanus tectorius* and *Phoenix reclinata*, several metabolites remained stable or showed only minor changes across sampling times. Based on this GCMS analysis, there is inconsistency of secondary metabolite accumulation in observed plants. Even though few exhibit patterns indicating changes in secondary metabolite content, however, the different types of secondary metabolite with a specific trend (such as increasing or decreasing) are not necessarily identical. Among the secondary metabolites detected, neophytadiene showed consistent responses across species (Fig. 7). Several plants, including *Agathis damara, Barringtonia asiatica, Dillenia indica, Inocarpus fagifer, Pandanus tectorius, Salacca zalacca,* and *Syzygium samarangense*, exhibited a steady increase in neophytadiene content from T0 to T2, suggesting that ALAN enhanced the accumulation of this compound over time. In other species, such as *Dalbergia cultrata, Diospyros blancoi, Parmentiera aculeata, and Phoenix reclinata,* levels remained relatively stable or showed moderate increases. By contrast, *Hanguana malayana* displayed a decline in neophytadiene at T2 after initially higher levels, while *Canarium indicum* and *Posoqueria latifolia* showed only minor variation across time points. Taken together, these results indicate that neophytadiene was one of the most responsive metabolites under ALAN, with several species showing progressive increases in its abundance. Since neophytadiene is a diterpenoid hydrocarbon often linked to plant defense and signaling, its accumulation under ALAN suggests a role in biochemical adjustment to nighttime light exposure.

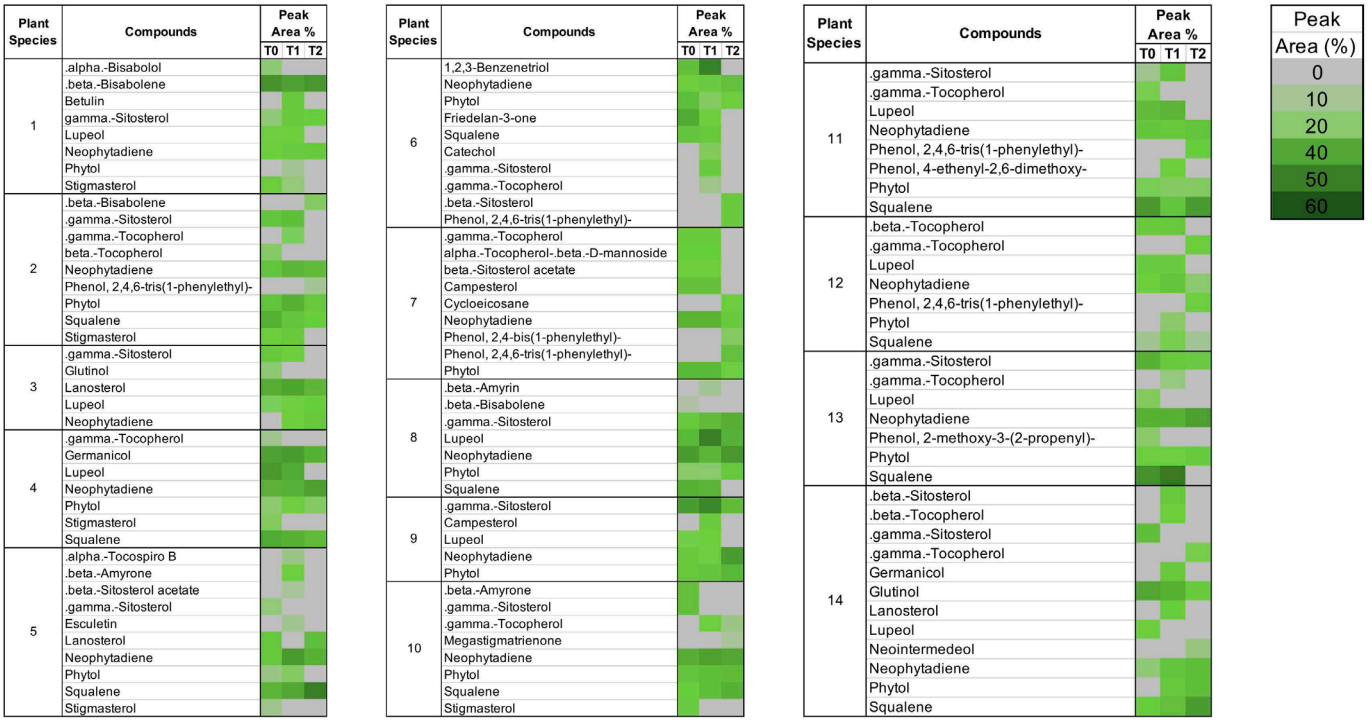

Fig. 6: Heatmap of SMs profiles from plants at BBG affected by ALAN, shows some of highest SM measured from, (a) *Agathis damara*, (b) *Barringtonia asiatica*, (c) *Canarium indicum*, (d) *Dalbergia cultrata*, (e) *Dillenia indica*, (f) *Diospyros blancoi*, (g) *Hanguana malayana*, (h) *Inocarpus fagifer*, (i) *Pandanus tectoris*, (j) *Parmentiera aculeata*, (k) *Phoenix reclinate*, (l) *Posoquera latifolia*, (m) *Salacca salacca*, (n) *Syzygium semarangense*.

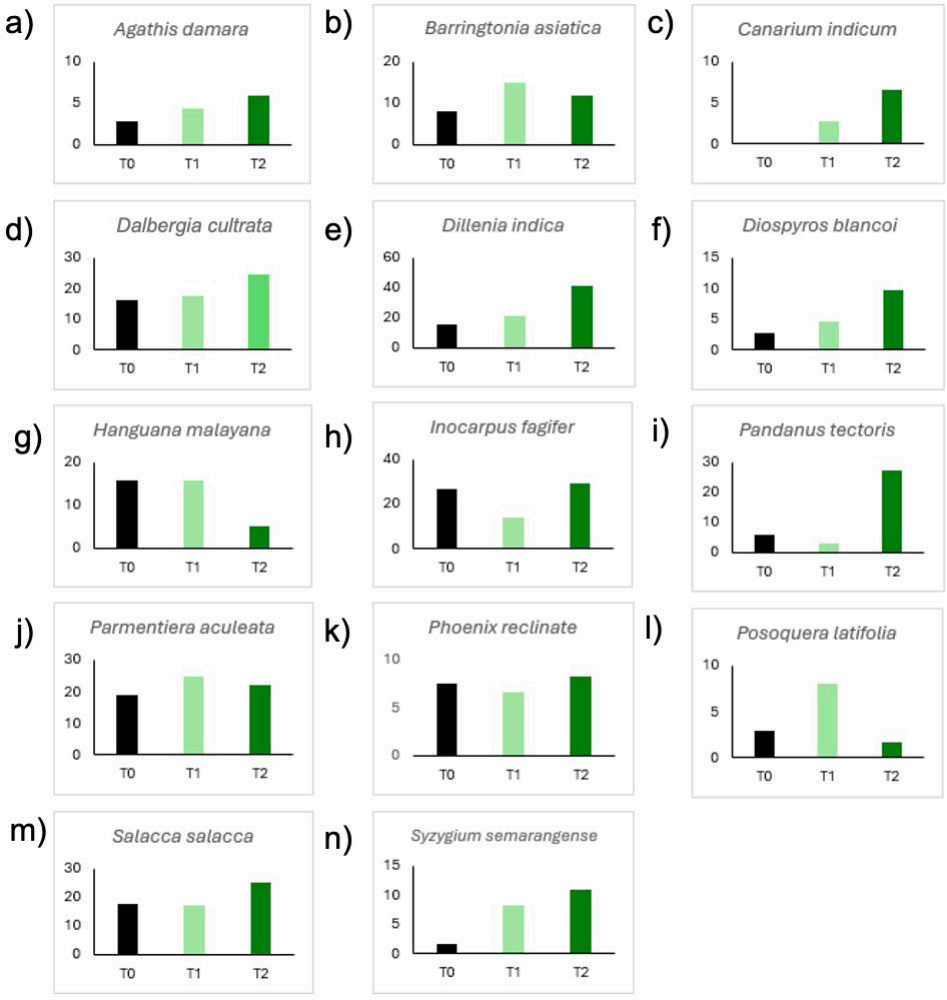

Fig. 7: Peak area of Neophytadiene using GC-MS in fourteen samples including, (a) *Agathis damara*, (b) *Barringtonia asiatica*, (c) *Canarium indicum*, (d) *Dalbergia cultrata*, (e) *Dillenia indica*, (f) *Diospyros blancoi*, (g) *Hanguana malayana*, (h) *Inocarpus fagifer*, (i) *Pandanus tectoris*, (j) *Parmentiera aculeata*, (k) *Phoenix reclinate*, (l) *Posoquera latifolia*, (m) *Salacca salacca*, (n) *Syzygium semarangense*.

Correlation

Correlations between *A*, secondary metabolite, and plant traits under ALAN are shown in Fig. 8. *A* were not strongly related to distance from the lamp (Fig. 8a) or plant growth form (Fig. 8b). However, *A* were positively associated with secondary metabolite changes at both T1 and T2 (Fig. 7c-d), suggesting that plants with greater accumulation of secondary metabolite tended to also maintain higher *A*. Secondary metabolite responses showed little association with distance from the lamp at either T1 or T2 (Fig. 8f, i), and growth form had minimal influence on distance placement (Fig. 8e). Nevertheless, growth form showed a slight effect on metabolite changes, with trees tending to accumulate more secondary metabolites compared to herbs and shrubs at both T1 and T2 (Fig. 8g, j). Furthermore, secondary metabolite changes at T1 were positively correlated with changes at T2 (Fig. 8h), indicating consistency in species' responses over time.

The content of plant mineral nutrients does not associate with the photosynthesis change of five plant species

Plant mineral nutrient analysis was conducted on five species, Four essential macronutrients - nitrogen (N), phosphorus (P), sulfur (S), and potassium (K) - which play important roles in plant stress tolerance, were examined. Results are depicted in Fig. 9. In addition, micronutrients such as chloride (Cl), which indirectly regulates stomatal movement, and iron (Fe), which is linked to photosynthesis and the activation of antioxidant synthesis, were also measured. Phosphorus (P) content generally showed an increasing trend across most species, except in *Pandanus tectorius*. Since phosphorus is essential for photosynthesis, metabolite production, and growth, its accumulation highlights its central role in plant physiology. However, in *Dalbergia cultrata*, the decline in *A* observed at T2 was not accompanied by a reduction in P, and similar patterns were observed in other species, indicating that changes in P content were not always directly associated with photosynthetic rates. With the exception of Fe, which displayed more consistent patterns, no uniform trend was observed across the mineral nutrients measured. Increases or decreases in nutrient levels varied among species, showing no consistent accumulation pattern after ALAN exposure. Interestingly, in species such as *Pandanus tectorius, Diospyros blancoi, Syzygium samarangense,* and *Parmentiera aculeata*, increases in *A* were accompanied by decreases in K content, with the exception of *Parmentiera aculeata*, which showed fluctuating K levels. Overall, the trends in mineral nutrient content did not consistently mirror the changes in photosynthetic rates observed in these species (Fig. 4).

Discussion

It is well known that light is one of the most important factors for photosynthesis in plants. ALAN altered photosynthetic rates indicating a species-specific effect. ALAN's exposure to plants in the BBG

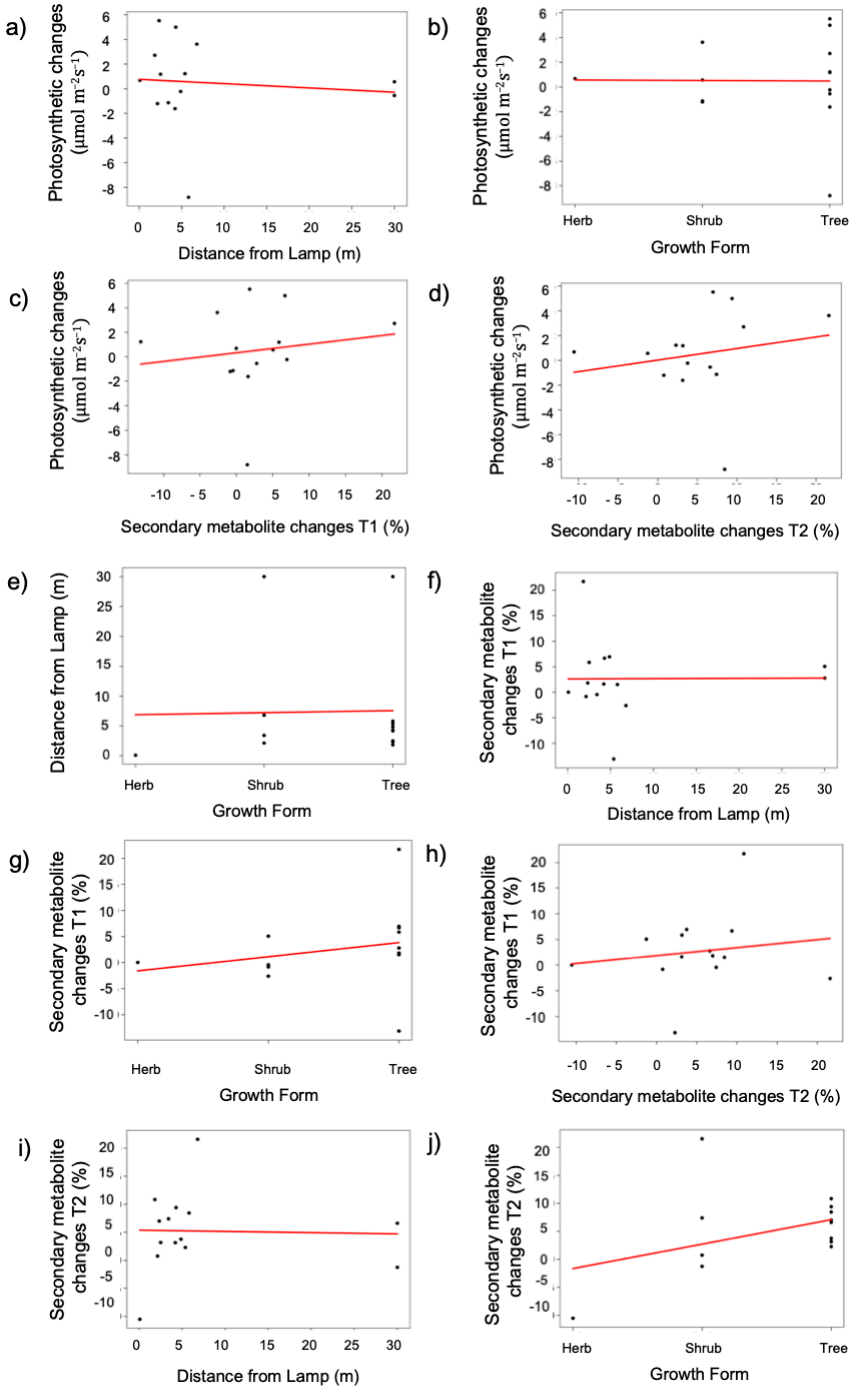

Fig. 8: Correlations between photosynthetic changes, secondary metabolites, and plant traits under ALAN. The correlation between (a) Photosynthesic changes and Distance from lamp, (b) Photosynthesic changes and growth form, (c) Photosynthesic changes and Secondary Metabolite changes at T1, (d) Photosynthesic changes and Secondary metabolite change at T2, (e) Distance from lamp and Growth form, (f) Secondary Metabolite changes at T1 and distance from lamp, (g) Secondary Metabolite changes at T1 and growth form, (h) Secondary Metabolite changes at T1and Secondary Metabolite changes at T2, (i) Secondary Metabolite changes at T2 and distance from lamp, and (j) Secondary Metabolite changes at T2 and growth form, from all fourteen species measured.

area was scheduled twice a week, for a maximum of 4 hours per day, beginning at around 6:00 PM when it started dark and there was no natural light. This suggests a potential of four hours long interruption in dark periods of the plants. Earlier research on *Cymbidium* plants grown under high Photosynthetic Photon Flux Density (PPFD) has revealed that along with reduced PSII activity, the photosynthetic rate increases during night interruption and decreases during the day (Kim et al., 2015). Earlier research demonstrates that plant physiological responses can be substantially altered by short durations of exposure or by dim light conditions during the night (Bennie et al., 2016; Singhal et al., 2019). Given that, we did not find a similar pattern from our result as a significant decrease of *A* after ALAN In *Dalbergia cultrata* and *Diospyros blancoi*, *A* was higher under ALAN at T1, but decrease at T2, suggesting that *A* efficiency may be reduced with longer exposure to ALAN. In contrast, *Syzygium samarangense* showed no changes in *A* under ALAN across both time measurement, reflecting a more stable of *A* under ALAN. In treatment plants, A was higher on Day 2 compared to Day 7, while control plants showed

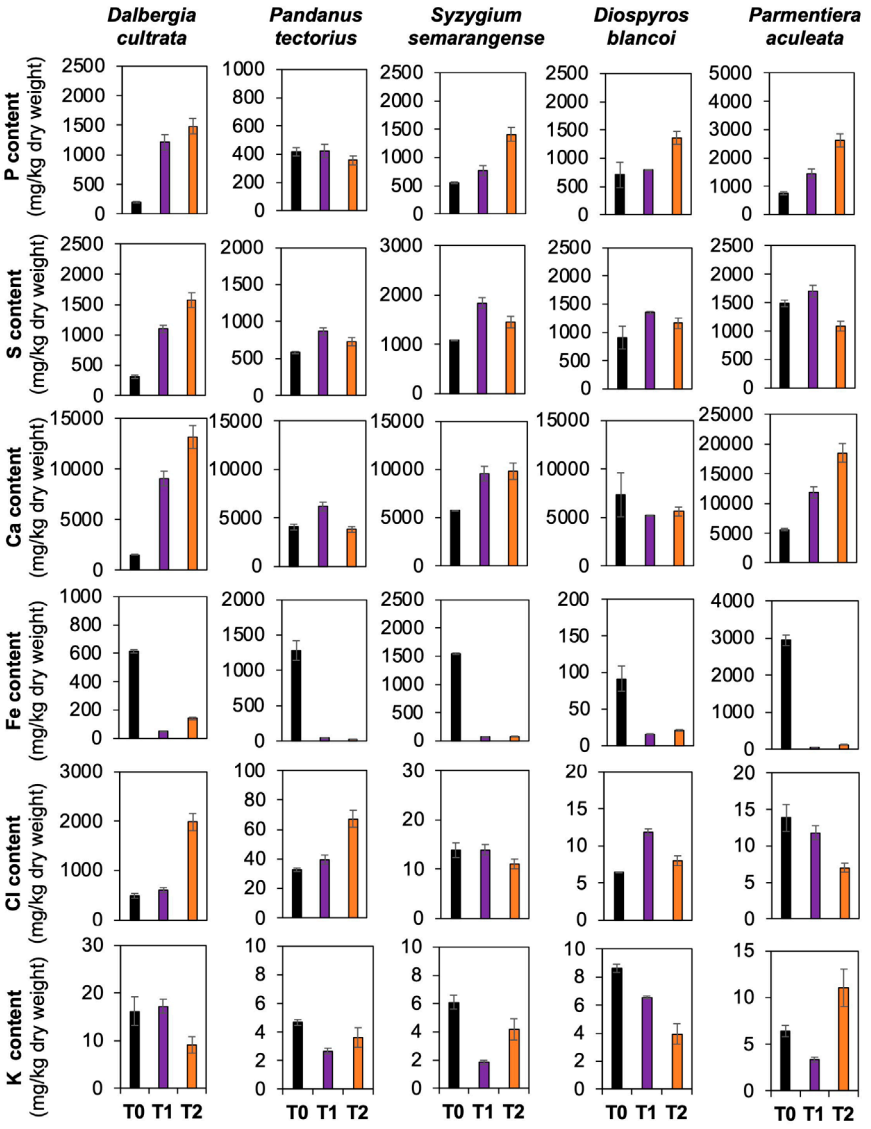

Fig. 9: Plant elemental contents of phosphorous (P), sulfur (S), calcium (Ca), iron (Fe), chloride (Cl), and potassium (K) in a.) *Dalbergia cultrata*, b.) *Pandanus tectorius*, c.) *Syzygium semarangense*, d.) *Diospyros borneensis*, and e.) *Parmentiera aculeata* leaves, before, 4 months, and 8 months after ALAN application (T0, T1, and T2, respectively). Values are means ± standard deviation (SD) from technical duplication.

no significant changes across time or intensity. Meanwhile, quite an opposite result was found from a study on *Poa pratensis* under low levels of ALAN which showed no significant change in photosynthesis affected by ALAN (Crump et al., 2021). Given the infrequent intensity of LED lamps installed and short brief switch-on periods at BBG, plants are unlikely to be regularly exposed to ALAN which reflects the variation of plants' response toward ALAN. Moreover, ALAN has a minimal impact on net photosynthesis in the environment due to outdoor lighting's lower quantum flux density compared to sunlight (Bennie et al., 2016).

Leaf gas exchange measurement shows g_s in *Dalbergia cultrata* and *Diospyros blanco*i, increased significantly at T1 under ALAN, but declined by T2. Stomata limitation is a significant barrier for photosynthesis and reduced g_s can lead to major limitation to photosynthesis under moderate stress (Wang et al., 2022). Previous studies also suggest a linear relationship between A and g_s, however, stomata can still respond independently to other factors such as environmental stress and may not directly affect the assimilation (Wong et al., 1979). Responses of g_s under ALAN were not uniform, with some species showing decrease while others showed increase across time measurement. Lower g_s in species such as *Dalbergia cultrata* and *Pandanus tectorius* suggests that plants may conserving the water under ALAN. In contrast, *Diospyros blancoi* and *Syzygium samarangense* appear to maintaining higher g_s under ALAN, which may support increased photosynthesis. Stomata aperture is a compromise between the need to conserve water and maintaining the rate of assimilation.

Studies within the field of ALAN have shown that secondary metabolite composition in plants, such as terpenoid, phenolic and alkaloid, can significantly change in response to nighttime light exposure (Flexas et al., 2012; Singhal et al., 2019). Several common compounds, including phytol, lupeol, squalene, sterols, and tocopherols, generally increased from T0 to T2. Among the studied species, *Diospyros blancoi* and *Syzygium samarangense* displayed a consistent increase, suggesting that these species are particularly responsive in their secondary metabolite production under ALAN. Neophytadiene is identified as volatile diterpene, it also has a role in response to abiotic stress (Zahratul et al., 2021; Zhou et al., 2020). Neophytadiene, consistently accumulated across most species, indicating that it may play an important role in protecting plants or mediating stress responses to ALAN. In contrast, *Hanguana malayana* showed a re-

duction of neophytadiene by T2, suggesting that its response was temporary rather than sustained. Previous study in *Brassia rapa* showed Neophytadiene was produced under all light conditions, where the highest Neophytadiene is found in the red-light treatment (14.76%) compared to natural light (2.47%). This indicates that the production of secondary metabolites, one of them is Neophytadiene, depends on the light treatment given. Moreover, its absence may affect the growth and survival in the environment (Saapilin et al., 2022). In our study, several secondary metabolites (*gamma-tocopherol, Neophytadiene, Stigmasterol and botulin*) related to abiotic stresses such as temperature, drought and salinity stresses were found in some plants (Abbasi et al., 2007; Du et al., 2022; Jafari Hajati et al., 2018; Zhang et al., 2023). Some of the light response-related secondary metabolites from our measurements such as *gamma-tocopherol* which is related to plant's stress signalling to light (Munné-Bosch, 2019), *gamma sitosterol* (Munné-Bosch, 2019) and *beta-sitosterol* (Terletskaya et al., 2021).

Light signaling and photosynthesis influence nutrient acquisition and utilization in plants (Xu et al., 2021). More specifically, phosphorus (P) has been shown to affect both photosynthesis and metabolite profiles in *Eucalyptus globulus* (Warren, 2011). However, in our study, P content did not show a clear association with changes in photosynthetic rate (*A*) across five species (*Dalbergia cultrata, Pandanus tectorius, Syzygium semarangense, Diospyros blancoi,* and *Parmentiera aculeata*). Although P is essential for ATP synthesis and energy transfer, its direct effect on carbon assimilation may be limited. Similarly, sulfur (S), which is a key constituent of many enzymes in the photosynthetic carbon reduction cycle, showed no consistent pattern with *A*, despite its known role in modulating photosynthetic efficiency. Potassium (K) deprivation is typically linked to reductions in photosynthetic efficiency (Erel et al., 2015), but in our analysis K trends also did not consistently match changes in photosynthesis.

Iron (Fe), on the other hand, showed a notable decrease in accumulation in the five tested species at T1 and T2 compared to T0. Since Fe is the primary transition metal in the protein complexes of the photosynthetic electron transport chain (Schmidt et al., 2020), it plays an essential role in supporting electron flow and chlorophyll biosynthesis. The observed reduction in Fe content after ALAN exposure may therefore not simply indicate reduction, but rather reflect increased Fe to sustain higher photosynthetic activity under altered light conditions. In this sense, ALAN may indirectly alter Fe allocation within plant tissues, highlighting its potential influence on nutrient dynamics even if its direct effect on photosynthetic rate was not always evident in our study.

Plant physiological responses to ALAN in the BBG area appear to be species-specific, as each species displayed a distinct trend in *A*, g_s, secondary metabolite and nutrient content. Light quality is another important factor, as different wavelengths (red, blue, and green) are absorbed differently and influence photosynthesis in distinct ways (Liu and Van Iersel, 2021).The differences observed between short-term and long-term exposure further suggest that ALAN effects may shift over time. For future experiments, species that showed consistent and strong responses across multiple parameters, such as *Diospyros blancoi* and *Syzygium samarangense*, could serve as good model species to investigate the mechanisms of ALAN tolerance and sensitivity. Moreover, secondary metabolite families such as diterpenoids (e.g., neophytadiene) or sterols, which were both strongly responsive to ALAN and correlated with photosynthetic changes, may provide a useful biochemical standard for monitoring plant responses. Long-term studies focusing on these species and metabolite families under controlled light conditions will be valuable to uncover the mechanistic links between ALAN, plant physiology, and ecological outcomes. From an applied perspective, the careful selection of urban tree species with higher ALAN tolerance, along with the use of light sources that minimize physiological disruption, will help reduce the ecological impacts of nighttime lighting while still supporting urban functions such as safety and tourism.

Conclusion

This study demonstrates that ALAN induces species-specific physiological changes in plants within the BBG area. We found that ALAN exposure changed *A* and g_s in several species, shifted accumulation patterns of secondary metabolites, and reduced the nutrient content across five species. While these changes were relatively small in magnitude and varied among species, the results clearly show that ALAN can impact various aspects of plant function. As modernization and urban development around the BBG area are inevitable, selecting plant species with higher tolerance to ALAN is crucial to minimising the ecological consequences of ALAN. Tree species such as *Diospyros blancoi* and *Syzygium samarangense*, which showed strong and consistent responses, may be good candidates for further research and management considerations. Further long-term studies are needed to fully understand the influence of ALAN on plant physiology and adaptation in natural ecosystems.

Acknowledgement

The authors would like to thank Deputy Research and Innovation, National Research and Innovation Agency for granting access permission to conduct the data collection in the Bogor Botanic Gardens area. We also thank Andrey Nugraha who helped us to collect the field data.

Funding

This work was supported by the Research Organization for Life Sciences and Environment, National Research and Innovation Agency. In the framework for the project Program House of Endangered Plant Conservation (DIPA-*Rumah Program Konservasi Tumbuhan Terancam Kepunahan*) with a grant number: 39/III.5/HK/2022

Conflict of interest

No potential conflict of interest was reported by the authors.

References

Abbasi, A.-R., Hajirezaei, M., Hofius, D., Sonnewald, U., Voll, L.M., 2007: Specific roles of α-and γ-tocopherol in abiotic stress responses of transgenic tobacco. Plant Phys. 143(4), 1720-1738. DOI: 10.1104/pp.106.094771

Ariati, S.R., Widyatmoko, D., 2019: Bogor botanic gardens. Sibbaldia: Int. J. Bot. Garden Horticult. 17, 11-28.

Bennie, J., Davies, T.W., Cruse, D., Gaston, K.J., 2016: Ecological effects of artificial light at night on wild plants. J. Ecol. 104(3), 611-620. DOI: 10.1111/1365-2745.12551

Crump, M.C., Brown, C., Griffin-Nolan, R.J., Angeloni, L., Lemoine, N.P., Seymoure, B.M., 2021: Effects of Low-Level Artificial Light at Night on Kentucky Bluegrass and an Introduced Herbivore. Front. Ecol. Evol. 9. DOI: 10.3389/fevo.2021.732959

Dalle Carbonare, L., Basile, A., Rindi, L., Bulleri, F., Hamedeh, H., Iacopino, S., Shukla, V., Weits, D.A., Lombardi, L., Sbrana, A., Benedetti-Cecchi, L., Giuntoli, B., Licausi, F., Maggi, E., 2023: Dim artificial light at night alters gene expression rhythms and growth in a key seagrass species (*Posidonia oceanica*). Scient. Reports 13(1), 10620. DOI: 10.1038/s41598-023-37261-3

Du, Y., Fu, X., Chu, Y., Wu, P., Liu, Y., Ma, L., Tian, H., Zhu, B., 2022: Biosynthesis and the roles of plant sterols in development and stress responses. Int. J. Mol. Sci. 23(4), 2332. DOI: 10.3390/ijms23042332

Erel, R., Yermiyahu, U., Ben-Gal, A., Dag, A., Shapira, O., Schwartz,

A., 2015: Modification of non-stomatal limitation and photoprotection due to K and Na nutrition of olive trees. J. Plant Physiol. 177, 1-10. DOI: 10.1016/j.jplph.2015.01.005

Fang, H., Qi, X., Li, Y., Yu, X., Xu, D., Liang, C., Li, W., Liu, X., 2020: De novo transcriptomic analysis of light-induced flavonoid pathway, transcription factors in the flower buds of *Lonicera japonica*. Trees 34(1), 267-283. DOI: 10.1007/s00468-019-01916-4

Farquhar, G.D., Sharkey, T.D., 1982: Stomatal Conductance and Photosynthesis. Annual Review Plant Physiol. 33(1), 317-345. DOI: 10.1146/annurev.pp.33.060182.001533

Flexas, J., Barbour, M.M., Brendel, O., Cabrera, H.M., Carriquí, M., Díaz-Espejo, A., Douthe, C., Dreyer, E., Ferrio, J.P., Gago, J., 2012: Mesophyll diffusion conductance to CO_2: an unappreciated central player in photosynthesis. Plant Sci. 193, 70-84. DOI: 10.1016/j.plantsci.2012.05.009

Fukuda, N., 2013: Advanced light control technologies in protected horticulture: A review of morphological and physiological responses in plants to light quality and its application. J. Develop. Sust. Agricult. 8(1), 32-40.

Gaston, K.J., Bennie, J., Davies, T.W., Hopkins, J., 2013: The ecological impacts of nighttime light pollution: a mechanistic appraisal. Biol. Reviews 88(4), 912-927. DOI: 10.1111/brv.12036

Jafari Hajati, R., Payamnoor, V., Ahmadian Chashmi, N., Ghasemi Bezdi, K., 2018: Improved accumulation of betulin and betulinic acid in cell suspension culture of *Betula pendula* roth by abiotic and biotic elicitors. Prep. Biochem. Biotechnol. 48(9), 867-876. DOI: 10.1080/10826068.2018.1514514

Kim, Y.J., Yu, D.J., Rho, H., Runkle, E.S., Lee, H.J., Kim, K.S., 2015: Photosynthetic changes in Cymbidium orchids grown under different intensities of night interruption lighting. Scientia Horticulturae 186, 124-128. DOI: 10.1016/j.scienta.2015.01.036

Liu, J., Van Iersel, M.W., 2021: Photosynthetic physiology of blue, green, and red light: Light intensity effects and underlying mechanisms. Front. Plant Sci. 12, 328. DOI: 10.3389/fpls.2021.619987

Munné-Bosch, S., 2019: Vitamin E Function in Stress Sensing and Signaling in Plants. Development Cell 48(3), 290-292. DOI: 10.1016/j.devcel.2019.01.023

Poulin, C., Bruyant, F., Laprise, M.-H., Cockshutt, A.M., Marie-Rose Vandenhecke, J., Huot, Y., 2014: The impact of light pollution on diel changes in the photophysiology of Microcystis aeruginosa. J. Plankton Res. 36(1), 286-291. DOI: 10.1093/plankt/fbt088

Saapilin, N.S., Yong, W.T.L., Cheong, B.E., Kamaruzaman, K.A., Rodrigues, K.F., 2022: Physiological and biochemical responses of Chinese cabbage (*Brassica rapa* var. *chinensis*) to different light treatments. Chem. Biol. Technol. Agric. 9(1), 27. DOI: 10.1186/s40538-022-00293-4

Sankhuan, D., Roytrakul, S., Nakano, M., Supaibulwatana, K., 2022: Proteomic sensing associated with terpenoid biosynthesis of *Artemisia annua* L. in response to different artificial light spectra. J. Plant Interact. 17(1), 19-32. DOI: 10.1080/17429145.2021.2009582

Schmidt, S.B., Eisenhut, M., Schneider, A., 2020: Chloroplast transition metal regulation for efficient photosynthesis. Trends Plant Sci. 25(8), 817-828. DOI: 10.1016/j.tplants.2020.03.003

Singhal, R.K., Kumar, M., Bose, B., 2019: Eco-physiological Responses of Artificial Night Light Pollution in Plants. Russ. J. Plant Physiol. 66(2), 190-202. DOI: 10.1134/S1021443719020134

Terletskaya, N.V, Korbozova, N.K., Kudrina, N.O., Kobylina, T.N., Kurmanbayeva, M.S., Meduntseva, N.D., Tolstikova, T.G., 2021: The influence of abiotic stress factors on the morphophysiological and phytochemical aspects of the acclimation of the plant *Rhodiola semenowii* Boriss. Plants 10(6), 1196.

Venkat, A., Muneer, S., 2022: Role of Circadian Rhythms in Major Plant Metabolic and Signaling Pathways. Front. Plant Sci. 13. DOI: 10.3389/fpls.2022.836244

Wang, Y., Wang, Y., Tang, Y., Zhu, X.-G., 2022: Stomata conductance as a goalkeeper for increased photosynthetic efficiency. Curr. Op. Plant Biol. 70, 102310. DOI: 10.1016/j.pbi.2022.102310

Warren, C.R., 2011: How does P affect photosynthesis and metabolite profiles of *Eucalyptus globulus*? Tree Physiol. 31(7), 727-739. DOI: 10.1093/treephys/tpr064

Wei, Y., Li, Z., Zhang, J., Hu, D., 2023: Effects of artificial light at night and drought on the photosynthesis and physiological traits of two urban plants. Front. Plant Sci. 14. DOI: 10.3389/fpls.2023.1263795

Weller, J.L., Kendrick, R.E., 2008: Photomorphogenesis and photoperiodism in plants. In: Photobiology: The Science of Life and Light, 417-463. Springer.

Wimalasekera, R., 2019: Effect of light intensity on photosynthesis. Photosynth, Product Environ. Stress, 65-73.

Wong, S.C., Cowan, I.R., Farquhar, G.D., 1979: Stomatal conductance correlates with photosynthetic capacity. Nature 282(5737), 424-426. DOI: 10.1038/282424a0

Xu, J., Guo, Z., Jiang, X., Ahammed, G.J., Zhou, Y., 2021: Light regulation of horticultural crop nutrient uptake and utilization. Hort. Plant J. 7(5), 367-379.

Zahratul, M., Muhamad, S., Wahyuning, A.S., 2021: Metabolomic analyses to evaluate the effect of drought stress on Chilli Pepper (*Capsicum annum* L.) at Vegetative Stage. Russ. J. Agric. Socio.-Ec. Sci. 111(3), 48-60. DOI: 10.18551/rjoas.2021-03.07

Zhang, D., Tang, X., Chen, L., Qiu, X., Song, C., Wang, H., Chang, Y., 2023: Functional characterization and transcriptional activity analysis of Dryopteris fragrans farnesyl diphosphate synthase genes. Front. Plant Sci. 14, 1105240. DOI: 10.3389/fpls.2023.1105240

Zhang, S., Zhang, L., Zou, H., Qiu, L., Zheng, Y., Yang, D., Wang, Y., 2021: Effects of Light on Secondary Metabolite Biosynthesis in Medicinal Plants. Front. Plant Sci. 12. DOI: 3389/fpls.2021.781236

Zhou, H.-C., Shamala, L.F., Yi, X.-K., Yan, Z., Wei, S., 2020: Analysis of terpene synthase family genes in *Camellia sinensis* with an emphasis on abiotic stress conditions. Sci Reports 10(1), 933. DOI: 10.1038/s41598-020-57805-1

ORCID
Mutiara K. Pitaloka https://orcid.org/0000-0001-5036-9189
Kartika Ning Tyas https://orcid.org/0000-0001-6196-9462
Muhayatun Santoso https://orcid.org/0000-0003-1623-7363
Isnaeni Isnaeni https://orcid.org/0000-0001-5749-9891
Andes Hamuraby Rozak https://orcid.org/0000-0001-9641-5830
Ahmad Fathoni https://orcid.org/0000-0002-2671-2407
Sukma Surya Kusumah https://orcid.org/0000-0001-5216-6246
Rizmoon N. Zulkarnaen https://orcid.org/0000-0001-6861-9641
Didi Usmadi https://orcid.org/0000-0002-9237-7133
Dwi Setyo Rini https://orcid.org/0000-0002-2792-7880
Triadiati Triadiati https://orcid.org/0000-0001-7845-9151
Maya Ismayati https://orcid.org/0000-0001-7651-1403
Frisca Damayanti https://orcid.org/0000-0001-5409-6640
Reza Ramdan Rivai https://orcid.org/0000-0002-8927-2429

Address of the corresponding author:
Mutiara K. Pitaloka, Research Center for Applied Botany, National Research and Innovation Agency, Cibinong, West Java, Indonesia.
E-mail: muti016@brin.go.id

Supplementary material

Tab. S1: Characteristic of plant species exposed to Artificial Light at Night (ALAN) and their habitat

No	Species	Family	Habitus	Type of lamp	Region	Distance of plant to lamp (m)
1	*Agathis dammara* (Lamb.) Rich. & A. Rich.	Araucariaceae	Tree	Circular spotlight with 38 nodes	B	4,20
2	*Barringtonia asiatica* (L.) Kurz.	Lecythidaceae	Tree	Circular spotlight with 18 nodes	A	4,86
3	*Canarium indicum*	Burseraceae	Tree	Circular spotlight with 56 nodes	B	30,00
4	*Dalbergia cultrata* Benth.	Leguminosae	Tree	Circular spotlight with 18 nodes	A	5,80
5	*Dillenia indica*	Dileniaceae	Tree	Circular spotlight with 18 nodes	A	1, 84
6	*Diospyros borneensis* Hiern	Ebenaceae	Tree	Circular spotlight with 18 nodes	A	2,33
7	*Hanguana malayana* (Jack) Merr.	Hanguanaceae	Aquatic herb	Reed shape LED Lamp	A	0,10
8	*Inocarpus fagifer* (Parkinson) Fosberg	Leguminosae	Tree	Circular spotlight with 18 nodes	A	5,37
9	*Pandanus tectorius* Parkinson ex Du Roi	Pandanaceae	Shrub	Circular spotlight with 18 nodes	A	1,35

Tab. S2: Leaf gas exchange measurment of plants at BBG

	E				Ci				VPDleaf				Tleaf			
	T1		T2		T1		T2		T1		T2		T1		T2	
Sampel	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment
Agathis damara	0,0012	0,0014	0,0009	0,0013	226,7039	228,0894	252,5949	298,0940	4,3415	4,2218	2,0314	3,9269	31,9377	31,5999	31,7432	33,0548
Barringtonia asiatica,	0,0013	0,0014	0,0023	0,0015	278,9274	305,4389	301,4918	271,6856	2,0180	1,8558	2,4829	1,9825	31,1534	30,0297	32,0355	31,0731
Canarium indicum	0,0014	0,0017	0,0013	0,0013	342,0721	323,7262	269,5905	292,0694	1,8921	1,9036	2,0990	1,9848	29,5564	30,6070	32,0651	31,2254
Dalbergia cultrata	0,0009	0,0029	0,0012	0,0011	294,5958	236,7322	263,9659	294,7054	2,0009	1,9625	1,9300	1,8242	30,9002	33,1250	30,9586	30,5914
Dillenia indica	0,0034	0,0022	0,0018	0,0023	379,3525	386,0560	325,9988	327,4864	1,7278	1,8222	1,9263	1,9260	29,7527	29,9461	31,3599	31,2009
Diospyros blancoi	0,0020	0,0109	0,0038	0,0030	423,8706	379,3310	352,2154	312,8772	1,8208	1,4386	1,8270	1,9304	30,2271	29,8990	30,4193	31,2790
Hanguana malayana	0,0034	0,0037	0,0016	0,0034	335,2215	336,6480	293,5427	324,2124	1,7511	2,0648	2,0369	2,0118	30,1558	32,5090	31,7767	32,0015
Inocarpus fagifer	0,0004	0,0007	0,0003	0,0003	285,4900	314,8010	268,8417	294,5174	2,0182	1,8992	3,0339	2,0218	30,9466	30,0113	30,1766	30,0015
Pandanus tectoris	0,0033	0,0024	0,0020	0,0025	258,0079	191,1727	309,2783	306,4333	3,8100	4,0154	1,8048	1,7852	34,0940	35,9668	30,0776	30,1471
Parmentiera aculeata	0,0020	0,0008	0,0027	0,0051	376,0995	369,0437	383,7985	368,1353	1,8399	1,8202	1,8249	1,2842	30,3961	30,2683	30,4033	30,2471
Phoenix reclinate	0,0023	0,0019	0,0024	0,0010	276,0175	339,2588	301,2802	292,7468	2,1317	2,2396	4,1572	1,9644	32,7650	30,0492	31,3256	31,4710
Posoquera latifolia	0,0024	0,0017	0,0018	0,0008	269,4933	273,1944	307,0918	249,6754	3,9890	4,2973	1,8841	2,1780	31,4654	32,1662	30,5101	32,5098
Salacca salacca	0,0015	0,0025	0,0006	0,0018	310,7811	290,3890	307,0432	278,1157	4,3037	4,1416	1,9791	4,0641	32,0216	32,1168	31,1887	31,0139
Syzygium semarangense	0,0007	0,0010	0,0008	0,0028	283,8591	292,3332	275,1648	295,6578	1,9821	1,9796	2,0119	1,9643	30,9597	30,7167	31,0633	31,4060