

Case Report

Artificial Light At Night (Alan) Influences Understory Plant Traits Through Ecological Processes: A Two-Year Experiment In A Rubber Plantation In China.

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Abstract

Nighttime artificial light (ALAN) revealed a novel ecological component that affects organisms in a variety of ways. However, nothing is known about how ALAN affects understory plants. Through a two-year field light experiment in a tropical rubber plantation in south China, we assessed whether ALAN would have an impact on the leaf mass per area (LMA) of understory plants. We postulated that ALAN might have two effects on the understory: directly increasing LMA by adding light to aboveground plant parts, and indirectly altering soil nutrient composition by drawing insects, which lowers LMA. Two species were chosen: *Melastoma candidum*, which represents species that require light, and *Colocasia gigantea*, which represents plants that can withstand shadow. We assessed soil nutrients, canopy openness, LMA, and each person's separation from light sources.

LMA and ALAN strength were shown to be negatively correlated in our Bayesian linear mixed model, suggesting that ALAN may have a more indirect effect on LMA by improving soil nutrient availability as opposed to directly functioning as a light resource. For *Colocasia gigantea*, this link was important, but not for *Melastoma candidum*. These findings imply that the effects of ALAN on the understory ecosystem may be intricate and species-specific. Our work emphasizes the necessity of ongoing investigation and knowledgeable anthropogenic ecosystem management.

Keywords : *Colocasia Gigantea; Melastoma Candidum; Artificial Light At Night; Functional Traits; Leaf Mass Per Area; Specific Leaf Area; Understory.*

INTRODUCTION

Since the early 20th century, artificial light at night (ALAN), one of the main causes of light pollution, has interfered with natural processes [1–3]. According to a recent study, light pollution has a negative impact on about 23% of the world's populated land areas, which represents more than 80% of the world's population [4]. ALAN affects the physiology or behavior of a wide range of taxonomic groups, including fish, amphibians, birds, mammals, invertebrates, plants, and amphibians [5–8]. This is true even though the intensity of ALAN varies by several orders of magnitude, from direct illumination of urban and suburban vegetation to faint skyglow reflected from distant cities [1].

Additionally, it might change how ecosystems function [5, 8]. ALAN, for instance, draws insects and obstructs their

development, reproduction, foraging, and mobility [9–11]. Insect population decreases have been connected to these interferences [9]. There aren't many published studies on how ALAN affects plants, in contrast to mammals [1,12,13]. Plant growth tests with and without weak ALAN (28 lux: within the range of light intensities at ground level under street lights) were carried out by Peiße et al. (2021), who demonstrated that ALAN enhances the biomass of herbaceous plants. According to those earlier investigations, plant biomass may be impacted by even low-intensity ALAN [9,13]. Few research, nevertheless, have looked at how ALAN affects plant functional features in settings that are similar to their natural habitat.

Plant leaf functional characteristics may be impacted by ALAN both directly and indirectly. First, because ALAN may function as an extra light source, it may have a direct impact on plant

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leaf functional characteristics like leaf mass per area (LMA). From low-cost, short-lived leaves to high-cost, long-lived leaves, LMA is the essential characteristic in the leaf economics spectrum [14–16]. Even though LMA is influenced by innate genetic processes [17], environmental factors including light, water, and temperature also have an impact. Through the use of photoreceptors, plants are able to perceive and react to four aspects of their light environment: duration, direction, intensity, and spectral quality of light [8,18].

According to Terashima et al. (2006) [19], sun leaves are thicker than shade leaves because their palisade tissue is higher. This is because the light-saturated rate of leaf photosynthesis per unit area is highly correlated with leaf structural parameters like leaf thickness, mass per area, mesophyll surface area, and chloroplast surface area. LMA for individual species was substantially higher in evergreen than deciduous species and proportional to species distributions along the insolation gradient [20–22]. Furthermore, Ackerly et al. (2002) showed that the average LMA values among a local community rose noticeably as potential diurnal insolation (PDI) increased.

However, because ALAN can attract insects and promote soil fertility, it may have an indirect effect on plant leaf functional features. Many insects are drawn to light and orient themselves by keeping a steady angle to light beams. According to earlier research, 30–40% of insects perish shortly after coming into contact with street lamps as a result of collision, scorching, dehydration, or predation [25, 26]. Dead insects killed by ALAN may be significant nutritional inputs for soil nutrients [28], as nitrogen (N) and phosphorus (P) are the nutrients that most commonly limit primary productivity in forest ecosystems [27]. LMA and leaf N and P contents are known to be influenced by soil resources, particularly N and P availability [16,29,30], and these effects are known to be contrary to those of light (i.e., intense light enhances LMA whereas high levels of N availability decrease LMA).

In this study, we used a two-year experiment in a tropical rubber plant forest in south China to examine the effects of ALAN on the LMA of understory plants. Anthropogenic ecosystems under land use can be represented by rubber plantations, one of the primary causes of deforestation in Southeast Asia [32], low canopy closure, and sparse shrub density. Ants are drawn to ALAN, according to a prior study carried out at this location using the identical experimental setup [11]. In order to identify variations in their reactions to ALAN, we chose two understory species as the experiment's participants, each of which represents a species that requires light and one that can withstand shade.

According to our theory, there are two different ways that ALAN would affect the understory plants. The first would be a direct effect, whereby ALAN increases LMA by acting as an additional light source for plants. The second route would be an indirect one, in which ALAN lowers LMA by increasing

soil nutrient availability. We calculated the effects of ALAN, daylight, and their interaction on the LMA of the understory plants in order to ascertain the relative significance of these two pathways.

MATERIALS AND METHODS

Experimental Setup

ALAN field experiments were conducted in a rubber tree forest (N21°54'0" E101°16'0"; elevation: 560 m;) in the Xishuangbanna Tropical Botanical Garden (XTBG), China. We set up five plots and chose two for this experiment based on the target plant species' natural distribution. The rubber trees were planted in rows, two meters apart, with a distance between each row varying from four to twelve meters.

While the understory has a significantly lower density than that of the core forest, the overstory is about 20 meters high. The research region experiences an average annual precipitation of 1557 mm and an average annual temperature of 21.7 °C. In the research area, no fertilizer was applied. At night, an artificial lighting environment was created in each plot using LED lights (10 W; Tc = 6696 K). The LED system was set up to run automatically every day from 8 p.m. until 5 a.m. The timing and length of the LED's nighttime activity were managed by an electric timer and a rechargeable lithium battery (12 V/30 Ah). About two meters above the ground, the LEDs and batteries were connected by an electric line and suspended from a tree branch with a lampshade. The LED bulbs at the light sources had a mean $\pm$ SE light intensity of 372.8 ± 66.6 lux 62.0 ± 4.9 lux, 190.2 ± 26.5 lux at a 1 m radius at a 2 m radius and 9.9 ± 0.7 lux at a 4 m radius. After 10 meters, the light intensity was hardly noticeable at 0 lux. Distance was therefore considered a continuous quantity. In order to capture the gradient effect of ALAN on plants in our investigation. Furthermore, it is possible to cognitively think of plant individuals farther away (beyond 5 m) as controls. Leaf disc samples were taken in November 2021, two years after the experiment was set up in November 2019.

Species Selection

When choosing species, we took into account the understory conditions, making sure that there were at least 15 adult individuals from the light source to ten meters away. The species' membership in functional groups—more especially, evergreen plants—and their absence from nitrogen-fixing families like Leguminosae were also crucial requirements. Therefore, we selected two species for our study, each from a separate plot: *Melastoma candidum*, which represents a species that requires light, and *Colocasia gigantea*, which represents a species that can withstand shadow.

Measurements

Selection of Pieces When choosing species, we took into account the understory conditions, making sure that there were at least 15 adult individuals from the light source to ten meters away. The species' membership in functional groups—more especially, evergreen plants—and their absence from nitrogen-fixing families like Leguminosae were also crucial requirements. Consequently, we selected two species for our investigation, each from a separate plot: *Colocasia*. Instead of utilizing whole-leaf LMA values, we employed a 1-cm-diameter punch to measure leaf edges and veins [3–7]. We selected between three and five healthy leaves from each individual *C. gigantea* plant and five healthy leaves from each individual *M. candidum* plant. We punched seven leaf discs from each leaf of *C. gigantea* and five leaf discs from each leaf of *M. candidum*. In total, we sampled 165 leaves from 35 *C. gigantea* individuals and 95 leaves from 19 *M. candidum* individuals. In order to evaluate the general trends in soil nutrients (N, C, and P), we collected surface soil samples in five plots in June 2019 and June 2022, ranging in depth from 0 to 10 cm. Three duplicates were taken at the location beneath the ALAN and 10 meters from the ALAN, independently from each plot. Following sampling, the soils were allowed to air dry at room temperature for a week before being sieved through 0.85 and 0.15 mm mesh. This was done in order to measure total N and C by combustion using an elemental analyzer (VarioMAXCN, Elemental Analysensysteme GmbH, Langelsbold, Germany) and total P by inductively coupled plasma atomic-emission spectrometer (iCAP7400, ThermoFisher Scientific, Waltham, MA, USA).

Data Analysis

For each species, we built a Bayesian linear mixed-effects model to examine the effects of ALAN, sunshine, and their interaction on *M. candidum* and *C. gigantea*. Each individual's leaf mass per area (LMA) was calculated as a function of their distance from the ALAN source, their canopy openness, and the relationship between their distance and canopy openness. Since the intensity of the ALAN decreases non-linearly with distance, we used a log and reciprocal function to translate the distance from the ALAN source for each participant. We then looked at each person's distance from the ALAN source for the ALAN effect and their canopy openness for the daylight effect. We included people as a random intercept in our models to account for the lack of independence among individuals of the same species.

The Hamiltonian Monte Carlo technique (HMC), which was implemented in Stan [3–8], was used to estimate posterior distributions and model parameters using weakly-informative priors [39]. With a convergence threshold of 1.1 [40] and effective sample sizes greater than 400 [41] for all parameters, the convergence of the posterior distribution was evaluated

using the Gelman-Rubin statistical test. R version 4.2.0 [42] was used for all statistical studies, and the R package targets version 1.2.0 for workflow management [43]. Codes are accessible on November 1, 2023.

RESULTS

The species was significantly impacted by the artificial light's influence at night (ALAN) and during the day on LMA. These effects were not significant for *Melastomacandidum*, but they were for *Colocasia gigantea*. ALAN caused a significant drop in the average individual LMA value in the instance of *C. gigantea* (slope: -0.1052 [95%CI: $-0.1500, -0.0613$]). *M. candidum* responded to ALAN with a comparable decrease in LMA value (slope: -0.0434 [95%CI: $-0.1147, 0.0278$]), but this change was not statistically significant. There were no significant effects for either species from the combination of the effects of ALAN and sunshine.

In all areas, independent of distance from ALAN, soil nutrients dropped from 2019 to 2022 [$X_{2022} / X_{2019} < 1$, where X indicates carbon (C), nitrogen (N), and phosphorus (P)]. However, the relative percentage change in each nutrient showed that, in the plot where *C. gigantea* was investigated, the locations under ALANs sustained more nutrients in 2022 ($C_{2022} / C_{2019} = 0.919, N_{2022} / N_{2019} = 0.948, P_{2022} / P_{2019} = 0.537$) than the locations 10m away from ALAN ($C_{2022} / C_{2019} = 0.663, N_{2022} / N_{2019} = 0.695, P_{2022} / P_{2019} = 0.492$). Less noticeable changes were identified between the places under ALAN ($C_{2022} / C_{2019} = 0.677, N_{2022} / N_{2019} = 0.761, P_{2022} / P_{2019} = 0.729$) and those 10 m distant from ALAN ($C_{2022} / C_{2019} = 0.712, N_{2022} / N_{2019} = 0.746, P_{2022} / P_{2019} = 0.742$) in the plot where *M. candidum* was investigated.

DISCUSSION

The concept that artificial light at night (ALAN) can induce leaf mass apescence (LMA) in understory plants is supported by our data; nevertheless, there are species-specific responses. The shade-tolerant species *Colocasia gigantea* showed a significant decrease in LMA at the commencement of ALAN, but not the light-demanding species *Melastomacandidum*. We go into further detail about the lessons learned from our experiment and the implications of the four findings for the impacts of ALAN below. From 2019 to 2022, we saw a noticeable decline in the nutritional content of the soil. This trend may be mainly explained by the lack of fertilizer inputs at our study site and the nutrient absorption traits of herubber trees [4–4]. Rubber trees are renowned for their strong nitrogen intake, which over time may result in noticeable nutrient depletion. This effect was obvious in our investigation, with both ALAN-influenced and non-influenced areas showing discernible declines in soil nutrient levels over

a two-year period. In the *C. gigantea*-populated plot, this reduction in soil nutrients appeared to be less noticeable close to ALAN. This pattern points to a possible beneficial effect of ALAN on soil nutrients, most likely due to insect attraction. ALAN is known to draw insects, which may contribute more nutrients to the soil, including N and P, as a result of collisions, warming, or predation [25, 26]. This ALAN-induced insect influx may help counteract the nutritional depletion of the intensive rubber trees, which helps to preserve soil nutrients when exposed to ALAN. Because sunshine increases LMA, the significant decrease in LMA for *C. gigantea* under ALAN may be mostly caused by indirect impacts mediated through changes in soil nutrient availability rather than direct effects of ALAN functioning as a light resource. Indirect impacts on LMA, as opposed to direct effects seen in the earlier study [13], may also result in higher plant growth rates, which could raise biomass. It's well recognized that the presence of ALAN attracts a higher density of insects, many of which may perish from collision, overheating, dehydration, or predation [25, 26]. The decomposition of these dead insects may result in a significant increase in soil nutrients, especially N and P [28, 45–47], which are essential for plant growth. Although more thorough research is needed to determine the exact number of dead insects and the resulting rise in soil nutrients, this increased nutrient availability may change *C. gigantea*'s source allocation strategy. Moreover, higher soil nutrient availability, especially N, may boost *C. gigantea*'s photosynthetic potential. A decrease in LMA may result from *C. gigantea* allocating more resources for rapid leaf growth rather than structural tissue because of the increased nutritional availability [14, 31]. However, ALAN had no discernible influence on LMA in the light-demanding species *M. candidum*. Light-demanding species are exposed to more light than shade-tolerant species, which could have a minor impact on their LMA. The influence of daylight was not significant however, therefore light-demanding species may naturally have lower intrinsic trait variability (ITV) than shade-tolerant species. More research is needed to examine the differences in ITV between species that require light and those that tolerate shade, even though numerous studies have examined ITV within various functional groups, such as plants and trees [4–8]. Furthermore, the lack of a significant change in LMA for *M. candidum* may be explained by the less noticeable soil nutrient variations between ALAN-influenced and non-influenced sections in the plot with *M. candidum* compared to the plot with *C. gigantea*. Because the ALAN treatment was applied to a slope in this plot, higher soil nutrients from dead insects drawn to the ALAN may be more likely to wash off and not remain in the soil [49–51]. On the other hand, ALAN's attraction to insects may be spatially dependent, resulting in some areas drawing fewer insects and hence experiencing a reduced soil fertilization impact. Even though our findings

partially validate our hypothesis, we also identify a number of areas that might use better and provide recommendations for further research. First, we restricted our investigation to a small number of species. In field experiments, gathering a large number of individuals from the same species is tough. The flexibility to include more species is provided by Bayesian hierarchical models. A limited dataset can be handled by taking species-level factors into account rather than treating each species independently [40]. More species should be included in future research to properly evaluate generality. Second, leaf-scale measurements need to be made more accurate and uniform. Some leaves may be shaded by their neighbor, which could lead to intermittent exposure to ALAN. Additionally, the leaf blade's angle affects light interception, therefore it should be included in assessments. To more accurately measure light interception at the leaf level, future research may need to take into account leaf-level cameras or light dataloggers. Third, we also acknowledge the significance of photosynthetic adjustment points and photosynthetic photon flux density (PPFD) [52] in shaping plant responses to light and overall photosynthetic performance. Future studies that use these measurements will offer a more thorough understanding of plant-ALAN interactions and adaptations to changing light conditions. We acknowledge that there are other significant qualities for resource acquisition and defensive tactics, even though our study concentrated on a single trait (i.e., LMA), which is the essential variable for the leaf economic spectrum. An increasingly common feature of the anthropogenic environment, ALAN can have intricate and subtle effects on ecosystems. While insolation, or natural light, has been shown to increase LMA [20, 21], our research suggests that ALAN may have an impact on LMA more indirectly by improving the availability of nutrients in the soil as opposed to acting as a light resource directly. ALAN may have an unforeseen effect on the productivity and health of forests by altering the functional characteristics of understory plants and possibly the dynamics of soil nutrients. Because many essential elements of field settings, like small nutrition changes, herbivores, and competitors, are typically lacking from controlled trials, they may underestimate species interactions and unpredictable environmental heterogeneity.

CONCLUSIONS

Artificial light at night (ALAN) is a growing issue that affects understory plants in a variety of ways and depending on the species. In this two-year field study, we looked at how two plant species—*C. gigantea* and *M. candidum*—reacted to ALAN in a southern Chinese rubber plantation. The LMA of these species showed varying responses, with *C. gigantea* being impacted by nutrient dynamics. These findings imply that understanding species-specific reactions and the

possible impacts of ALAN on nutrient dynamics are crucial. Our work emphasizes the necessity of ongoing investigation and knowledgeable management of artificial ecosystems created by humans.

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