Artificial light at night advances reproductive phenology and reduces reproductive capacity of a wild plant

Shuo Wang[1], Zhihui Wang[2], Lu Xiao[1], Hongxiang Zhang[1], Yanjie Liu[1*]

[1]*Key Laboratory of Wetland Ecology and Environment, State Key Laboratory of Black Soils Conservation and Utilization, Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun, 130102, China*

[2]*Heilongjiang Bayi Agricultural University, Daqing, 163319, China*

*Email of corresponding author: liuyanjie@iga.ac.cn +86 431 82536096

Running title: light pollution effects on wild plant

Abstract

With the rapid increase in artificial light at night (ALAN) during the last century, a growing proportion of the world's ecosystems have been exposed to ALAN. While the effects of low-intensity ALAN on morphology and phenology are well known in laboratory and greenhouse studies, little is known about these effects of ALAN on wild plants. Moreover, how these morphological and phenological responses affect the fitness of wild plants is unclear.

Using the annual herbaceous plant, *Elsholtzia densa*, as a model species, we performed a field manipulation experiment and tested the responses of plant morphology, reproductive phenology, and reproductive capacity to ALAN.

We found that ALAN significantly increased the specific leaf area but decreased the number of secondary branches. It also advanced the onset of budding, blooming, fruiting, and seed maturity from 3.4 to 6.2 d. In addition, ALAN significantly increased the length of the top inflorescences of the species but decreased the percentage of the individuals with undeveloped top inflorescences. Consequently, ALAN reduced the number of fruity inflorescences.

Our results demonstrated that low-intensity ALAN, even with a relatively short period of exposure (i.e., one growing season), could cause passive responses in morphological traits of the wild plant, resulting in a decline in its reproductive capacity. In addition, the advanced reproductive phenology (e.g., flowering and blooming) induced by ALAN may cause plant-pollinator mismatches and threaten species coexistence at the community level.

Keywords: competition, community composition, light pollution, plant-environment interaction, population dynamics, species diversity, trophic level

Introduction

To adapt to the cyclical changes in the environmental light caused by the Earth's rotation, a wide range of organisms (e.g., algae, plants, and animals) have evolved molecular circadian clocks (Yerushalmi & Green, 2009; Hölker et al., 2010), which reduced competition and increased coexistence among sympatric species (Yerushalmi & Green, 2009; Gaston et al., 2013; Krittika & Yadav, 2020; Patke et al., 2020; Zhao et al., 2021). However, the Earth's dark night, which has existed for hundreds of millions of years, has rapidly disappeared because of the extensive use of artificial light at night (ALAN) in the past few decades (Falchi et al., 2016; Kyba et al., 2017; Gaston, 2018; Irwin, 2018). Clearly, such rapid global increases in ALAN can have serious impacts on various species and ultimately be a threat to ecosystems (Irwin, 2018). Therefore, testing the ecological effects of ALAN on organisms has become a trending topic in global change biology (Davies & Smyth, 2018; Hölker et al., 2021).

A recent meta-analysis indicated that most studies on the impacts of ALAN have focused on the behavior, physiology, and life history of animals (Sanders et al., 2021). Plants that can directly use light as a resource and source of information to regulate their growth and development (Gaston et al., 2015; Bennie et al., 2016; Singhal et al., 2019) are also likely to be affected by ALAN. Indeed, it is well-established that even a short duration or a low-intensity ALAN could markedly affect plant physiology (Smith 1982; Bennie et al., 2016). Moreover, many cases of urban trees, ornamental garden plants, and agricultural crops also show that ALAN can cause marked changes in plant physiological processes, morphology, and phenology (Liao et al., 2014; Haque et al., 2015; Kim et al., 2015; Bennie et al., 2016; Singhal et al., 2019). Surprisingly, except for a few studies (Bennie et al., 2018; Speißer et al., 2021; Liu et al., 2022), the unintended ecological impacts of ALAN on natural vegetation, such as wild plants, have rarely been tested (Bennie et al., 2016).

Given the importance of phenotypic plasticity in plant environmental tolerance (Sultan 2000; Richards et al., 2006; Liu et al., 2016), testing plastic responses of important functional traits to ALAN could help understand how wild plants adapt to such environmental changes. Recent studies have found that low-intensity ALAN, such as street lighting, could induce a photosynthetic response in plants (Speißer et al., 2021) and, thus, affect the growth and resource allocation of vascular plants (Speißer et al., 2021) and algae (Diamantopoulou et al., 2021). According to the hypothesis proposed by Briggs (2006), low-intensity ALAN with a varied spectral composition compared to that of direct sunlight could be interpreted as a shaded environment by plants. Consequently, plants may respond to ALAN by adjusting a suite of functional traits, such as maximum height and specific leaf area to avoid or tolerate such 'shading' conditions (Liu et al., 2016). However, the potential effects of ALAN on functional traits, especially leaf function and resource-management strategies of wild plants, have been poorly investigated.

In addition to its effects on functional traits, ALAN can also influence plant phenology. For example, both studies analyzing satellite data (Zheng et al., 2021) and phenology observations from the USA National Phenology Network (Meng et al., 2022) show that ALAN could advance the start date of the growing season. In contrast, the effects of ALAN on the flowering of ornamental plants could be negative, neutral, or positive (Cathay & Campbell, 1975). Such varied effects of ALAN on flowering have also been observed in wild plants (Bennie et al., 2018). Consequently, whether and how ALAN influences plant flowering and other reproductive phenology remains an open question, although changes in such phenology may have a significant effect on plant fitness and, thus, population dynamics. In addition, Knop et al. (2017) showed for the first time that ALAN could indirectly reduce plant reproductive success (i.e., decrease the fruit set of plants) by disrupting nocturnal pollination networks. Because such fitness consequences are essential for species coexistence

within a community, ALAN may exhibit profound effects on population and community dynamics (Bennie et al., 2015; Gaston et al., 2015; Bennie et al., 2018; Senzaki et al., 2020). However, to date, no study has tested the direct effects of ALAN on the fitness of wild plants.

In this study, we experimentally manipulated ALAN in a grassland and examined its effects on the morphology, reproductive phenology, and reproductive capacity of the widespread annual herbaceous species, *Elsholtzia densa* Benth. (Labiatae). We answered the following questions: 1) Does ALAN affect the morphological traits of the wild species? 2) Does ALAN affect the reproductive phenology of the wild species? 3) Does ALAN affect the reproductive capacity of the wild species?

Materials and methods

Study sites and species

This study was an additional experiment of a long-term grassland ALAN-manipulation experiment in a grassland in Inner Mongolia, China (50.17°N, 119.39°E, 523 m a.s.l.). The grassland has a temperate continental monsoon climate with a mean annual precipitation of 375 mm and a mean annual temperature of –3 °C (Liu et al., 2021). The large experiment is consisted of 40 square plots, each with an area of 4 m^2, which included four combinations of two levels of light-pollution (ALAN vs. no-ALAN) and two levels of animal-exclusion (open cage vs closed cage) treatments (10 replicates of each treatment combination). To homogenize the species composition of the 40 plots (Fig. 1), we tilled the soil and removed clonal propagules of species in each plot and then re-established artificial communities on April 18, 2021, with seeds of 54 common herbaceous species collected from the nearby grasslands in autumn 2020. To test how ALAN affects the morphology, reproductive phenology, and reproductive capacity of the wild plant species, the present study was conducted based on the 20 plots within the large experiment (Fig. 1), which were all treated

with open cages (i.e., no animal exclusion) but manipulated with two levels of light-pollution treatments (ALAN vs no-ALAN). The study species was an annual herbaceous species, *E. densa*, which belongs to the Lamiaceae family. According to the online database POWO (https://powo.science.kew.org), this species is widely distributed from East Afghanistan to Southeast Siberia and China. It was also the dominant species in the local communities of the 20 experimental plots, with a relative cover of 33.78 ± 3.0% in 2021 (Fig. S1).

Manipulation of the Artificial-light-at-night (ALAN)

The ALAN treatment was achieved by installing LED spotlights (LED Flood Light, 10 W, DC 12–24 V, IP 66, cool white 6500 K; Shenzhen Mingkeming Lighting Co., Ltd., China), which emit photosynthetically active radiation (Fig. S2), at a height of 2 m above the center of each plot using metal frames. To reduce the lateral light radiation, each of the 10 LED spotlights was covered with a cubic metal lampshade (30 × 30 × 30 cm). To avoid light interference between plots, the distance between two adjacent plots was 2 or 3 m. To simulate the light intensity caused by light pollution at ground level, two layers of white nylon nets and one layer of black light shading nets were used to cover the cubic metal lampshade. It resulted in a light intensity of 21.15 ± 0.30 lx, which falls into the range of ground-level intensity under streetlights (Bennie et al., 2016; Speißer et al., 2021). The lamps for the ALAN treatment were controlled by a photoelectric switch (XT KG-F, Zhejiang Xintuo New Energy Co., Ltd., China), automatically turning on/off when the ambient light intensity was below/above 30 lx, respectively. For the control level of light-pollution treatment (i.e., no-ALAN), we also installed the metal frames and lamp shades covered with nylon nets but without LED spotlights.

Trait measurements

From July 6 to September 23, 2021, we measured 14 traits (Table S1) related to morphology, reproductive phenology, and reproductive capacity. Before flowering (i.e., on July 6, 2021), we randomly selected and marked ten individuals of the study species in a 1 × 1 m subplot at the center of each plot. From July 6 to the starting date of the frost (i.e., September 23, 2021), we monitored the onset of budding, blooming, fruiting, and seed maturity of each *E. densa* individual daily for a total of 80 d. The onset of budding was defined as the presence of the first bud, blooming as the first purple inflorescence, fruiting as the first swelling inflorescence, and seed maturity as the first dehiscent inflorescence (similar approach see Dorji et al., 2013). We also measured the branch number and plant height for each of the marked individuals at the end of the growing season (i.e., on September 23). Branches arising from the main stem were defined as the primary branches and those arising from the primary branches as the secondary branches (similar approach see Li et al., 2016). To assess the responses of plant reproductive capacity to ALAN, we additionally measured the fruity inflorescence number (the number of inflorescences bearing seeds) for each marked individual. In addition, we recorded the number of individuals with undeveloped top inflorescences (i.e., individuals with top inflorescences staying at the bud stage until the end of the growing season) among the 10 marked individuals in each plot. Because we noticed that the top inflorescence of the study species seemed to be longer in ALAN plots than that in no-ALAN plots (Fig. 1c), we also measured the length of the top inflorescence to confirm such a promotional effect. Based on these data, we then calculated the percentage of individuals with undeveloped top inflorescences in each plot (the number of individuals with undeveloped top inflorescence / 10 marked individuals per plot × 100%).

To test the response of specific leaf area to ALAN, we collected ten fully expanded and healthy mature leaves from five unmarked individuals (i.e., two leaves per individual) next to

the subplot in each plot. We first scanned total leaf areas of the ten leaves from each plot and dried them at 60 ℃ for 72 h. After weighing the dry biomass of the scanned leaves, the specific leaf area (leaf area/leaf biomass) was calculated for each plot. To measure the total biomass and the aboveground and inflorescence biomass fractions of the study species under each treatment combination, we also randomly selected an additional 3–6 individuals adjacent to the subplot of each plot, resulting in a total of 45 individuals for ALAN plots vs. 52 individuals for no-ALAN plots. We harvested belowground parts, inflorescences, and other aboveground parts separately for each of the 97 individuals during the vigorous plant growth stage in late August. All materials were dried at 60 ℃ for 72 h and weighed. Based on the obtained biomass data, we calculated the total biomass (belowground parts + inflorescences + other aboveground parts), aboveground biomass fraction ([inflorescences + other aboveground parts]) / total biomass), and inflorescence biomass fraction (inflorescences / total biomass) for each individual.

Statistical analyses

For all 14 traits, we fitted Bayesian models using the 'brm' function with the '*brms*' package (version 2.14.4; Bürkner, 2017) in R (version 4.0.2; R Core Team, 2020). For each model, the response variable was the specific trait. All models included ALAN as the fixed effect. Except for the model for specific leaf area and percentage of individuals with undeveloped top inflorescences in each plot, we also included the identity of the plot as a random effect to account for non-independent repeated measurements. The number of fruity inflorescences and primary and secondary branches were modeled with negative binomial distributions, and the percentage of individuals with undeveloped top inflorescences was modeled with binomial distribution. Plant height and total biomass were modeled with gamma distributions, and the aboveground and inflorescence biomass fractions were modeled with beta

distributions. The remaining traits, including specific leaf area (square root transformed), budding date, blooming date, fruiting date, seed maturity date, and length of top inflorescence, were reasonably approximated to Gaussian error distributions after assessment by posterior predictive checks. The treatment ALAN was considered significant when the 95% credible interval of the posterior distribution did not overlap zero and as marginally significant when the 90% credible interval did not overlap zero.

Results

Effects of ALAN on morphological traits

Three of the six morphological traits were affected by ALAN. Specifically, ALAN significantly increased the specific leaf area (+9.2%; Fig. 2a; Table 1) but decreased the number of secondary branches (−44.3%; Fig. 2c; Table 1) compared to the no-ALAN treatment. Relative to the no-ALAN treatment, ALAN also tended to increase the aboveground biomass fraction (+0.68%; Fig. 2f; Table 1).

Effects of ALAN on reproductive phenology

ALAN strongly affected the reproductive phenology of *E. densa* (Table 1). Specifically, ALAN advanced the onset of budding, blooming, fruiting, and seed maturity by 3.4, 6.2, 3.4, and 4.6 d, respectively (Fig. 3; Table 1).

Effects of ALAN on reproductive capacity

Except for the inflorescence biomass fraction, the other three traits related to reproductive capacity were also affected by ALAN (Table 1). Specifically, ALAN significantly increased the length of the top inflorescences of the species (+29.17%; Fig. 4a; Table 1) but decreased the percentage of the individuals with undeveloped top inflorescences (−81.8%; Fig. 4b;

Table 1). Moreover, ALAN tended to reduce the number of fruity inflorescences (−25.2%; Fig. 4d; Table 1) relative to the no-ALAN treatment.

Discussion

To better understand the ecological effects of artificial lighting on population and/or community dynamics, it is necessary to link them to the responses of individual traits, especially those closely related to plant fitness. Using the widespread annual herbaceous plant *E. densa* as a model species, we found detrimental effects of ALAN on the sexual reproductive capacity of plants, which may be related to its effects on the morphological traits and reproductive phenology of *E. densa*.

Effects of ALAN on morphological traits

As mentioned above, low-intensity ALAN, relative to the direct sunlight, could be likened to a shaded environment for plants (Briggs, 2006) and, thus, could induce plant responses similar to shade-tolerance or shade-avoidance. The present study showed that the wild plant *E. densa* produces thinner and relatively larger leaves, which may enhance light capture per gram of leaf tissue and result in high mass-based photosynthesis. In addition, the plants allocated more biomass to the aboveground parts in response to ALAN. This is also a typical shade-tolerance-like response that increases the light capture efficiency of plants (Gommers et al., 2013). In this study, we observed a shade-avoidance-like response, showing that the wild plant produced fewer secondary branches in response to ALAN. Regardless of these shade-tolerance-like or shade-avoidance-like responses to ALAN, it could lead to a higher ability of light capture in day and consequently vigorous vegetation growth of the wild plant during the growth season (Gommers et al., 2013). However, the decrease in the number of secondary branches induced a lower fitness (i.e., fewer inflorescences) of the plant species.

This can partly reflect the fact that the secondary branches are the main organ for inflorescence production in the wild plant species (Shuo Wang, personal observation). Our study highlighted that plant morphological plasticity in response to ALAN is not necessarily adaptive (i.e., it increases fitness or maintains fitness homeostasis). The plastic changes we found might simply reflect the passive responses of the wild plant to ALAN.

Effects of ALAN on reproductive phenology

Because of photoperiodism (Garner & Allard, 1922), it is predicted that ALAN could influence flowering phenology in wild plants (Bennie et al., 2016). Surprisingly, our results showed that ALAN has not only driven the earlier flowering of the wild plant but also the earlier onset of blooming, fruiting, and seed maturity. This may be because the study species *E. densa* is a long-day plant. Based on our personal observations, the species, under ambient light conditions, generally starts its bud differentiation in mid-July, when the day length is approximately 15.8 h in the study region. Another study by Bennie et al. (2018) that was conducted at a similar latitude (approximately 50°N) also found that ALAN could cause earlier flowering in two of the three dominant grass species in a semi-grassland. Given that latitude is an important factor in determining plant photoperiod (Ettinger et al., 2021), future studies should test how ALAN generally influences plant flowering, as well as other reproductive phenology across latitudes. However, Bennie et al. (2018) also indicated that ALAN does not consistently cause earlier flowering for all species in a community. Such varied influences on flowering among different species may consequently result in plant-pollinator mismatches and, thus, further threaten pollination and plant reproduction (Forrest, 2015; Kudo & Cooper, 2019). Therefore, ALAN may also indirectly affect plant-pollinator interactions by altering plant phenology, in addition to their direct disruption of plant-pollinator interactions by negatively affecting nocturnal and diurnal pollinator communities

(Knop et al., 2017; Macgregor et al., 2017; Giavi et al., 2021).

Effects of ALAN on reproductive capacity

Compared to the ambient light condition (i.e., no-ALAN treatment), we found that ALAN increased the length of the top inflorescence but decreased the number of undeveloped-top-inflorescence individuals in each plot. This indicates that ALAN can promote the development and dimensions of the top inflorescence of the wild plant. However, better development of the top inflorescence may result in fewer side branches due to apical dominance (Beveridge et al., 2009; Domagalska & Leyser, 2011; Barbier et al., 2017; Kebrom, 2017). Indeed, our results showed that ALAN reduced the number of secondary branches and fruity inflorescences, indicating that the reproductive capacity of the wild plant decreased in response to ALAN. Although the plant allocated more biomass to the aboveground parts, the individuals under ALAN conditions showed similar reproductive allocation and even fewer fruity inflorescences than those under the no-ALAN condition. This implies that the additional biomass allocated to the aboveground parts was not used to increase reproduction and highlights the importance of linking fitness consequences with changes in functional traits in the context of light pollution.

Conclusions

We simulated and measured the effects of ALAN on the morphology, reproductive phenology, and reproductive capacity of the wild plant species *E. densa* under realistic field conditions. Our results demonstrated that low-intensity ALAN, within a relatively short period (i.e., one growing season), could cause passive responses in morphological traits of *E. densa*, resulting in a decline in its reproductive capacity. In addition, ALAN also advanced reproductive phenology (e.g., flowering and blooming) of the wild plant, which may lead to

plant-pollinator mismatches and threaten species coexistence. Because *E. densa* is an important nectar plant (Jiang et al., 1996), a substantial decline in the number of inflorescences with increasing ALAN may cause food shortages for some insects, such as bees, and, thus, affect ecosystem services via a bottom-up effect.

Acknowledgements

We thank the Erguna Forest-Steppe Ecotone Ecosystem Research Station, Institute of Applied Ecology, Chinese Academy of Sciences for providing the experimental field site and practical assistance with the experiment setup. We thank Xue Zhang, Zhengkuan Lu for their help in experiment setup. YL was supported by the Chinese Academy of Sciences (Y9B7041001). HZ was supported by the National Natural Science Foundation of China (41971069).

Author contributions

YL conceived the idea and designed the long-term grassland ALAN-manipulation experiment. SW and YL conceived the idea of present experiment and designed it. SW, ZW and LX performed the experiment. SW and YL analyzed the data. SW wrote the first draft of the manuscript, with major inputs from YL, and further inputs from ZW, LX and HZ.

Data accessibility

Should the manuscript be accepted, the data supporting the results will be archived in Dryad and the data DOI will be included at the end of the article.

References

Barbier FF, Dun EA, Beveridge CA. 2017. Apical dominance. Current Biology, 27(17):R864-R865.

Bennie J, Davies TW, Cruse D, Bell F, Gaston KJ. 2018. Artificial light at night alters grassland vegetation species composition and phenology. Journal of Applied Ecology, 55(1):442-450.

Bennie J, Davies TW, Cruse D, Gaston KJ. 2016. Ecological effects of artificial light at night on wild plants. Journal of Ecology, 104(3):611-620.

Bennie J, Davies TW, Cruse D, Inger R, Gaston KJ. 2015. Cascading effects of artificial light at night: resource-mediated control of herbivores in a grassland ecosystem. Philosophical Transactions of the Royal Society B: Biological Sciences, 370(1667):20140131.

Beveridge CA, Dun EA, Rameau C. 2009. Pea has its tendrils in branching discoveries spanning a century from auxin to strigolactones. Plant Physiology, 151(3):985-990.

Briggs W (2006). Physiology of plant responses to artificial lighting. Ecological consequences of artifical night lighting. C. Rich and T. Longcore, Island Press: 389-411.

Bürkner P. 2017. brms: An R Package for Bayesian Multilevel Models Using Stan. Journal of Statistical Software, 80(1):1 - 28.

Cathay AR, Campbell LE. 1975. Effectiveness of five vision-lighting sources on photo-regulation of 22 species of ornamental plants. Journal of the American Society for Horticultural Science, 100, 65-71.

Davies TW, Smyth T. 2018. Why artificial light at night should be a focus for global change research in the 21st century. Global Change Biology, 24(3): 872-882.

Diamantopoulou C, Christoforou E, Dominoni DM, Kaiserli E, Czyzewski J, Mirzai N, Spatharis S. 2021. Wavelength-dependent effects of artificial light at night on

phytoplankton growth and community structure. Proceedings of the Royal Society B: Biological Sciences, 288(1953): 20210525.

Domagalska MA, Leyser O. 2011. Signal integration in the control of shoot branching. Nature Reviews Molecular Cell Biology, 12(4):211-221.

Dorji T, Totland Ø, Moe SR, Hopping KA, Pan J, Klein JA. 2013. Plant functional traits mediate reproductive phenology and success in response to experimental warming and snow addition in Tibet. Global Change Biology, 19(2):459-472.

Ettinger AK, Buonaiuto DM, Chamberlain CJ, Morales-Castilla I, Wolkovich EM. 2021. Spatial and temporal shifts in photoperiod with climate change. New Phytologist, 230: 462-474.

Falchi F, Cinzano P, Duriscoe D, Kyba CCM, Elvidge CD, Baugh K, Portnov BA, Rybnikova NA, Furgoni R. 2016. The new world atlas of artificial night sky brightness. Science Advances, 2(6):e1600377.

Forrest JRK. 2015. Plant-pollinator interactions and phenological change: what can we learn about climate impacts from experiments and observations? Oikos, 124: 4-13.

Garner WW, Allard HA. 1922. Photoperiodism, the response of the plant to relative length of day and night. Science, 55, 582-583.

Gaston KJ. 2018. Lighting up the nighttime. Science, 362(6416):744-746.

Gaston KJ, Bennie J, Davies TW, Hopkins J. 2013. The ecological impacts of nighttime light pollution: a mechanistic appraisal. Biological Reviews, 88(4):912-927.

Gaston KJ, Visser ME, Hölker F. 2015. The biological impacts of artificial light at night: the research challenge. Philosophical Transactions of the Royal Society B: Biological Sciences, 370(1667):20140133.

Giavi S, Fontaine C, Knop E. 2021. Impact of artificial light at night on diurnal plant-pollinator interactions. Nature Communications, 12:1690.

Gommers CMM, Visser EJW, Onge KRS, Voesenek LACJ, Pierik R. 2013. Shade tolerance: when growing tall is not an option. Trends in Plant Science, 18(2): 65-71.

Haque MS, Kjaer KH, Rosenqvist E, Ottosen C. 2015. Continuous light increases growth, daily carbon gain, antioxidants, and alters carbohydrate metabolism in a cultivated and a wild tomato species. Frontiers in Plant Science, 6:522.

Hölker F, Bolliger J, Davies TW, Giavi S, Jechow A, Kalinkat G, Longcore T, Spoelstra K, Tidau S, Visser ME, Knop E. 2021. 11 pressing research questions on how light pollution affects biodiversity. Frontiers in Ecology and Evolution, 9:767177.

Hölker F, Wolter C, Perkin EK, Tockner K. 2010. Light pollution as a biodiversity threat. Trends in Ecology & Evolution, 25(12):681-682.

Irwin A. 2018. The dark side of light: how artificial lighting is harming the natural world. Nature, 553(7688):268-271.

Jiang Y, Deng Y, Yang W, Dang R. 1996. Anatomic studies of the floral nectaries in *Elsholtzia densa* Benth. Acta Botanica Boreali-Occidentalia Sinica, 16(3):239-244.

Kebrom TH. 2017. A growing stem inhibits bud outgrowth — the overlooked theory of apical dominance. Frontiers in Plant Science, 8:1874.

Kim YJ, Yu DJ, Rho H, Runkle ES, Lee HJ, Kim KS. 2015. Photosynthetic changes in *Cymbidium* orchids grown under different intensities of night interruption lighting. Scientia Horticulturae, 186:124-128.

Knop E, Zoller L, Ryser R, Gerpe C, Hörler M, Fontaine C. 2017. Artificial light at night as a new threat to pollination. Nature, 548(7666):206-209.

Kudo G, Cooper EJ. 2019. When spring ephemerals fail to meet pollinators: mechanism of phenological mismatch and its impact on plant reproduction. Proceedings of the Royal Society B: Biological Sciences, 286(1904): 20190573.

Krittika S, Yadav P. 2020. Circadian clocks: an overview on its adaptive significance. Biological Rhythm Research, 51(7):1109-1132.

Kyba CCM, Kuester T, De Migue AS, Baugh K, Jechow A, Hölker F, Bennie J, Elvidge CD, Gaston KJ, Guanter L. 2017. Artificially lit surface of Earth at night increasing in radiance and extent. Science Advances, 3(11):e1701528.

Li F, Chen B, Xu K, Gao G, Yan G, Qiao J, Li J, Li H, Li L, Xiao X, Zhang T, Nishio T, Wu X. 2016. A genome-wide association study of plant height and primary branch number in rapeseed (Brassica napus). Plant Sci, 242(169-177).

Liao Y, Suzuki K, Yu W, Zhuang D, Takai Y, Ogasawara R, Shimazu T, Fukui H. 2014. Night break effect of LED light with different wavelengths on floral bud differentiation of *Chrysanthemum morifolium* ramat 'Jimba' and 'Iwa no hakusen'. Environmental Control in Biology, 52(1):45-50.

Liu H, Wang R, Lü X, Cai J, Feng X, Yang G, Li H, Zhang Y, Han X, Jiang Y. 2021. Effects of nitrogen addition on plant-soil micronutrients vary with nitrogen form and mowing management in a meadow steppe. Environmental Pollution, 289:117969.

Liu Y, Dawson W, Prati D, Haeuser E, Feng Y, van Kleunen M. 2016. Does greater specific leaf area plasticity help plants to maintain a high performance when shaded? Annals of Botany, mcw180.

Liu Y, Speißer B, Knop E, van Kleunen M. 2022. The Matthew effect: Common species become more common and rare ones become more rare in response to artificial light at night. Global Change Biology,

Macgregor CJ, Evans DM, Fox R, Pocock MJO. 2017. The dark side of street lighting: impacts on moths and evidence for the disruption of nocturnal pollen transport. Global Change Biology, 23(2):697-707.

Meng L, Zhou Y, Román MO, Stokes EC, Wang Z, Asrar GR, Mao J, Richardson AD, Gu L, Wang Y. 2022. Artificial light at night: an underappreciated effect on phenology of deciduous woody plants. PNAS Nexus, 1(2): pgac046.

Patke A, Young MW, Axelrod S. 2020. Molecular mechanisms and physiological importance of circadian rhythms. Nature Reviews Molecular Cell Biology, 21(2):67-84.

R Core Team. 2020. R: A language and environment for statistical computing, R Foundation for Statistical Computing Vienna, Austria.

Richards CL, Bossdorf O, Muth NZ, Gurevitch J, Pigliucci M. 2006. Jack of all trades, master of some? On the role of phenotypic plasticity in plant invasions. Ecology Letters, 9: 981-993.

Sanders D, Frago E, Kehoe R, Patterson C, Gaston KJ. 2021. A meta-analysis of biological impacts of artificial light at night. Nature Ecology & Evolution, 5, 74-81.

Senzaki M, Barber JR, Phillips JN, Carter NH, Cooper CB, Ditmer MA, Fristrup KM, McClure CJW, Mennitt DJ, Tyrrell LP, Vukomanovic J, Wilson AA, Francis CD. 2020. Sensory pollutants alter bird phenology and fitness across a continent. Nature, 587(7835):605-609.

Singhal RK, Kumar M, Bose B. 2019. Eco-physiological responses of artificial night light pollution in plants. Russian Journal of Plant Physiology, 66(2):190-202.

Smith H. 1982. Light quality, photoperception and plant strategy. Annual Review of Plant Physiology, 33, 481-518.

Speißer B, Liu Y, van Kleunen M. 2021. Biomass responses of widely and less-widely naturalized alien plants to artificial light at night. Journal of Ecology, 109(4):1819-1827.

Sultan SE. 2000. Phenotypic plasticity for plant development, function and life history. Trends in Plant Science, 5: 537-542.

Yerushalmi S, Green RM. 2009. Evidence for the adaptive significance of circadian rhythms. Ecology Letters, 12(9):970-981.

Zhao K, Ma B, Xu Y, Stirling E, Xu J. 2021. Light exposure mediates circadian rhythms of rhizosphere microbial communities. ISME Journal, 15(9):2655-2664.

Zheng Q, Teo HC, Koh LP. 2021. Artificial light at night advances spring phenology in the United States. Remote Sensing, 13(3):399.

Table 1. Output of Bayesian models testing the effects of artificial light at night (ALAN) on each of the 14 traits observed in this study. Model estimates of ALAN whose 95% credible intervals do not overlap with zero are indicated with asterisks (*) and those whose 90% credible intervals do not overlap with zero are indicated with daggers (†).

No.	Trait	brm family	Term	Fixed effect			Random effect
				Estimate	95% CIs	90% CIs	Estimate (95% CIs)
1	*Specific leaf area*	Gaussian	Intercept *	14.087	(13.707, 14.473)	(13.767, 14.404)	—
	(*n* = 20)		ALAN *	0.630	(0.067, 1.181)	(0.171, 1.084)	—
2	*Primary branch number*	Negative binomial	Intercept *	2.132	(2.009, 2.252)	(2.03, 2.232)	0.14 (0.04, 0.25)
	(*n* = 200)		ALAN	0.060	(-0.110, 0.237)	(-0.08, 0.203)	—
3	*Secondary branch number*	Negative binomial	Intercept *	1.192	(0.763, 1.659)	(0.832, 1.575)	0.26 (0.01, 0.78)
	(*n* = 200)		ALAN *	-0.637	(-1.322, -0.006)	(-1.183, -0.111)	—
4	*Aboveground biomass fraction*	Beta	Intercept *	2.375	(2.32, 2.44)	(2.326, 2.425)	—
	(*n* = 97)		ALAN †	0.081	(-0.007, 0.170)	(0.007, 0.155)	—
5	*Height*	Gamma	Intercept *	0.024	(0.021, 0.027)	(0.021, 0.026)	0.01 (0.00, 0.02)
	(*n* = 200)		ALAN	0.001	(-0.004, 0.004)	(-0.003, 0.003)	—
6	*Total biomass*	Gamma	Intercept *	0.330	(0.234, 0.438)	(0.25, 0.418)	0.13 (0.08, 0.21)
	(*n* = 97)		ALAN	0.006	(-0.139, 0.153)	(-0.114, 0.124)	—
7	*Budding date*	Gaussian	Intercept *	205.513	(204.369, 206.629)	(204.575, 206.446)	0.49 (0.02, 1.35)
	(*n* = 200)		ALAN *	-3.588	(-4.954, -2.243)	(-4.713, -2.464)	—
8	*Blooming date*	Gaussian	Intercept *	229.190	(228.101, 230.243)	(228.313, 230.059)	1.29 (0.39, 2.46)
	(*n* = 200)		ALAN *	-5.494	(-7.673, -3.457)	(-7.294, -3.785)	—
9	*Fruiting date*	Gaussian	Intercept *	238.063	(237.033, 239.111)	(237.216, 238.896)	0.96 (0.09, 2.07)
	(*n* = 200)		ALAN *	-3.335	(-5.107, -1.583)	(-4.814, -1.885)	—
10	*Seed maturity date*	Gaussian	Intercept *	255.622	(254.520, 256.745)	(254.722, 256.548)	0.81 (0.04, 2.12)
	(*n* = 200)		ALAN *	-4.526	(-6.609, -2.462)	(-6.259, -2.784)	—
11	*Length of top inflorescence*	Gaussian	Intercept *	5.373	(4.942, 5.798)	(5.015, 5.728)	0.23 (0.01, 0.60)
	(*n* = 200)		ALAN *	0.836	(0.255, 1.429)	(0.348, 1.329)	—
12	*Percentage of individuals with undeveloped top inflorescence*	Binomial	Intercept	-0.060	(-0.191, 0.070)	(-0.171, 0.049)	—
	(*n* = 20)		ALAN *	-1.449	(-1.682, -1.218)	(-1.645, -1.257)	—
13	*Inflorescence biomass fraction*	Beta	Intercept *	-1.809	(-2.015, -1.610)	(-1.977, -1.645)	0.22 (0.04, 0.41)
	(*n* = 97)		ALAN	0.020	(-0.264, 0.299)	(-0.215, 0.254)	—
14	*Fruity inflorescence number*	Negative binomial	Intercept *	3.208	(2.966, 3.446)	(3.007, 3.404)	0.32 (0.17, 0.51)
	(*n* = 200)		ALAN †	-0.285	(-0.628, -0.050)	(-0.567, -0.008)	—

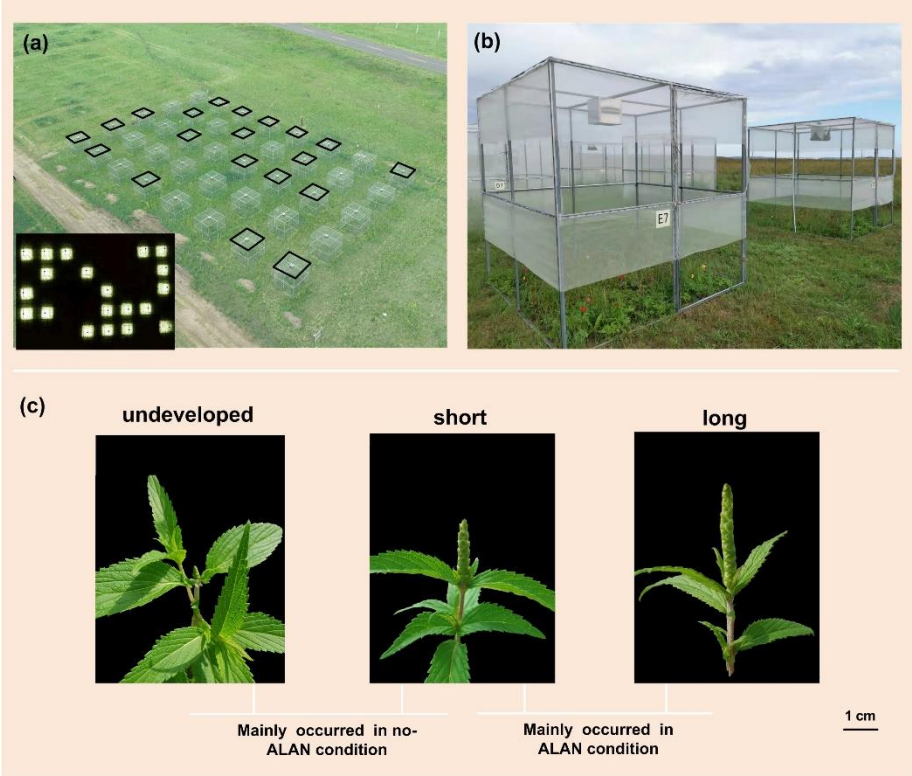

Figure 1. (a) Top view of the entire experiment during the day and night (lower left corner of the graph) (the current study was conducted in 20 open cages [i.e., no animal exclusion plots] marked with black borders). (b) Photograph of the plot without animal exclusion (i.e., open cage). (c) Morphological variation of top inflorescence among individuals under different artificial light at night (ALAN) treatments (left, undeveloped inflorescence; middle, short top inflorescence; right, long top inflorescence).

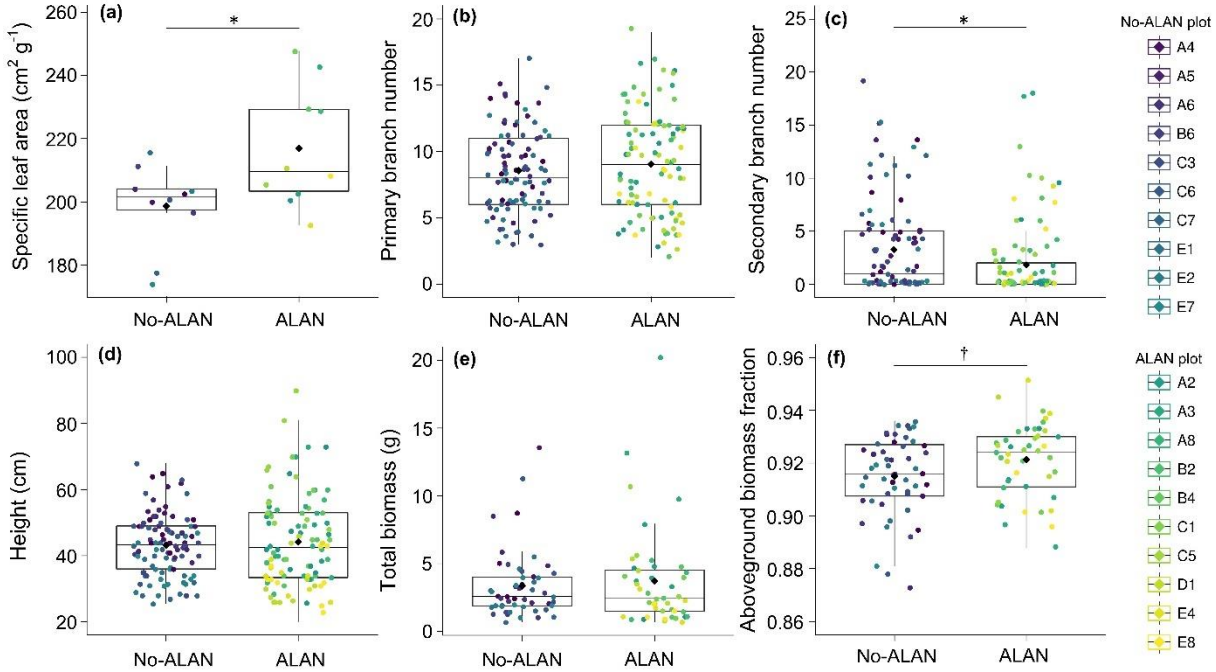

Figure 2. Effects of artificial light at night (ALAN) on specific leaf area (a), primary branch number (b), secondary branch number (c), plant height (d), total biomass (e), and aboveground biomass fraction (f). Asterisks (*) indicate the 95% credible intervals of model estimates of ALAN that do not overlap zero, and dagger (†) represents the 90% credible intervals do not overlap zero. For details on the sample size of each trait, see Table S1.

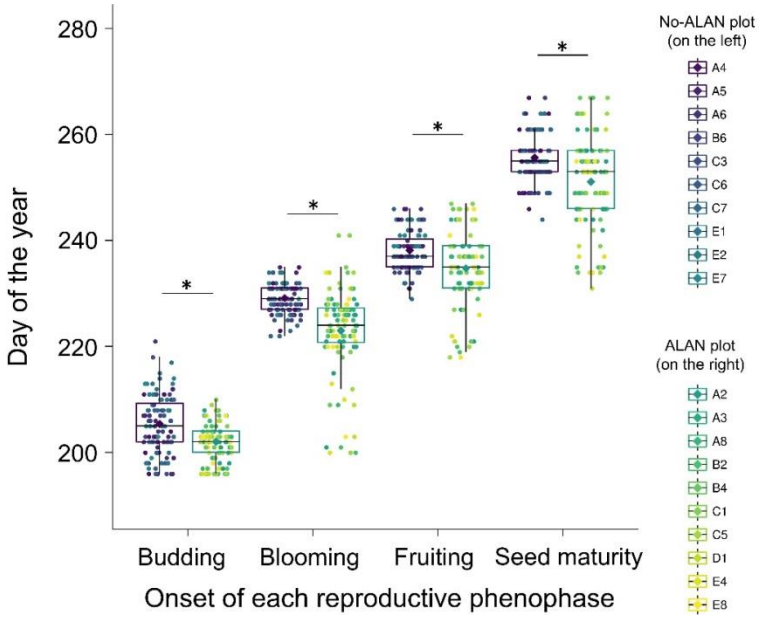

Figure 3. Effects of artificial light at night (ALAN) on the onset of different reproductive phenophases (expressed as Julian day of year 2021). Asterisks (*) indicate the 95% credible intervals of model estimates of ALAN that do not overlap zero. For details on the sample size of each trait, see Table S1.

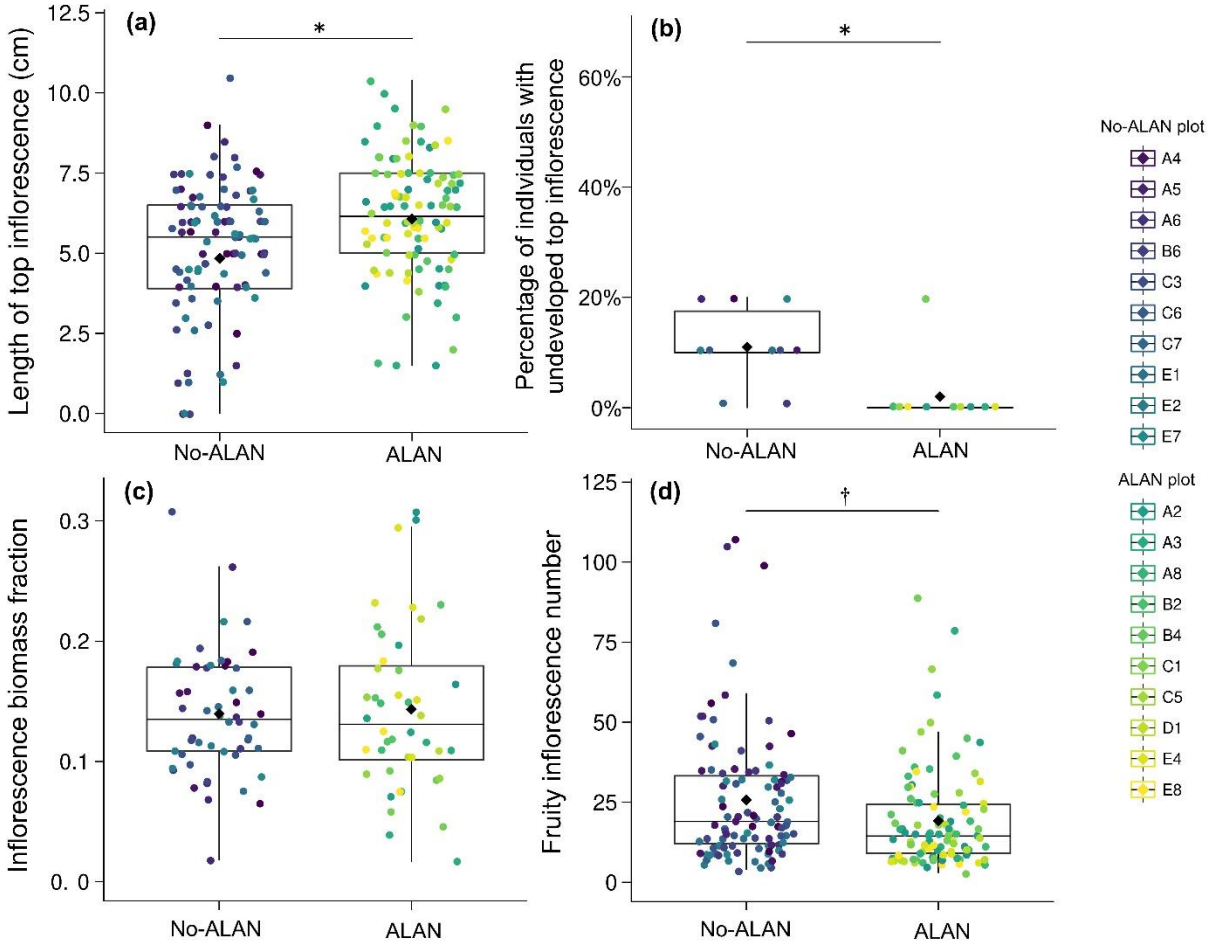

Figure 4. Effects of artificial light at night (ALAN) on length of top inflorescence (a), percentage of individuals with undeveloped top inflorescence (b), inflorescence biomass fraction (c), and fruity inflorescence number (d). Asterisks (*) indicate the 95% credible intervals of model estimate of ALAN that do not overlap zero, and daggers (†) represent the 90% credible intervals that do not overlap zero. For details on the sample size of each trait, see Table S1.

Supporting information

Table S1. The 14 traits measured in this study.

Category	Trait	Measuring time	Replication No.
Morphology	*Specific leaf area*	August 27, 2021	20
	Primary branch number	September 23, 2021	200
	Secondary branch number	September 23, 2021	200
	Aboveground biomass fraction	August 27, 2021	97
	Height	September 23, 2021	200
	Total biomass	August 27, 2021	97
Reproductive phenology	Budding date	From July 6 to	200
	Blooming date	September 23, 2021	200
	Fruiting date	(before plant budding	200
	Seed maturity date	to the start of frost)	200
Reproduction capacity	*Length of top inflorescence*	September 23, 2021	200
	Percentage of individuals with undeveloped top inflorescence	September 23, 2021	20
	Inflorescence biomass fraction	August 27, 2021	97
	Fruity inflorescence number	September 23, 2021	200

At the vigorous plant growth stage, August 27, 2021; the end of the growing season (start of frost), September 23, 2021.

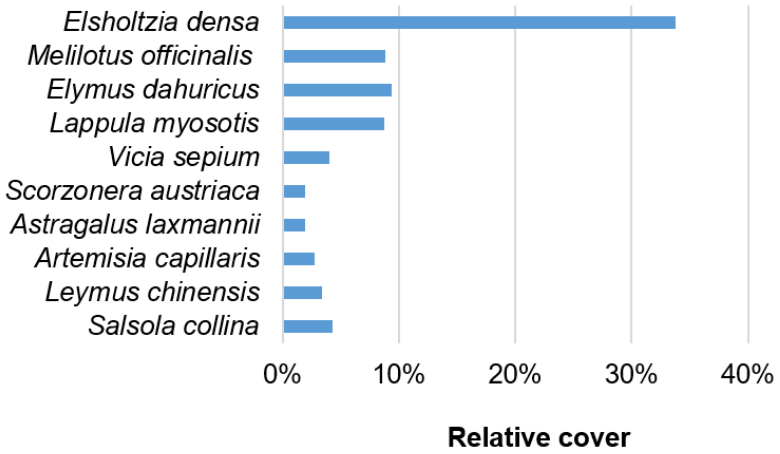

Figure S1. Top ten species ranked by the relative cover in 2021.

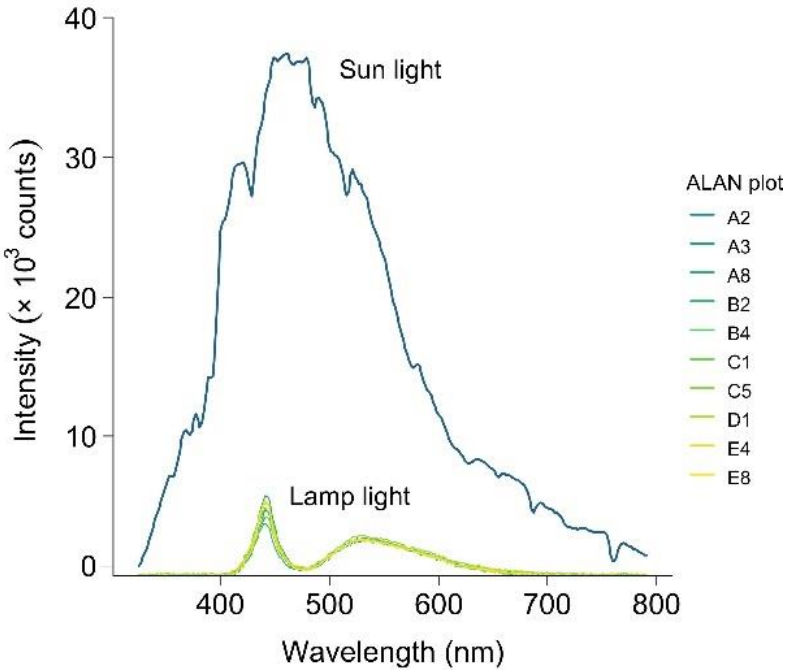

Figure S2. Spectral composition of sunlight (dark blue line) and the artificial light emitted by the ten LED spotlights (other lines, colored by plot). The photosynthetically active radiation ranges from 400 to 700 nm.