Seasonal and diurnal lag effects in chlorophyll fluorescence response of Acer campestre to heat stress

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Research Article

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Abstract

This study uses chlorophyll fluorescence metrics to explore the seasonal and diurnal lag effects in the heat stress response of Acer campestre, a common urban tree species in the UK. A two-phase design was employed: a controlled mesocosm experiment in 2022 and in situ field monitoring in 2023. In 2022, time series and lag analysis of NPQ and ETRmax revealed significant delays in physiological response during morning hours. NPQ exhibited lag effects at 0, -1, and − 2 days, with a significant negative association between same-day leaf temperature and NPQ (β = -0.03, $p = 0.026$), indicating reduced heat dissipation under thermal stress. ETRmax demonstrated a significant anticipatory decline two days before peak temperatures (β = -0.08, $p = 0.046$), suggesting downregulation of photosynthesis as a pre-emptive protective mechanism. In contrast, evening data in 2022 displayed no significant lag effects, indicating more synchronised physiological responses. During the 2023 season, field-based light response curves showed strong diurnal patterns without lag. ETRmax increased progressively from 14.84 at 09:30 am to 21.92 at 04:30 pm ($p < 0.001$), indicating increasing photosynthetic capacity throughout the day. NPQ peaked in early July but showed no statistically significant correlation with leaf temperature at any time point. These findings highlight marked differences in physiological behaviour between morning and evening and across years with different thermal intensities. Morning lag effects were especially pronounced in 2022, suggesting a “thermal memory” where previous heat exposure alters current physiological responses. These insights underscore the importance of accounting for time-of-day and delayed stress effects when evaluating urban tree resilience and developing adaptive urban greening strategies under intensifying climate extremes.

Introduction

Urban trees play a key role in meeting the United Nations’ Sustainable Development Goals (SDGs) by reducing the urban heat island effect and regulating microclimates, which improves thermal comfort (Turner-Skoff and Cavender 2019; Meier and Scherer 2012; Kaplan 1984; Coutts et al. 2015; Armson et al. 2012). However, heat extremes have become more frequent, severe, and prolonged across the globe, leading to significant environmental, economic, and public health crises (IPCC, 2023), and are projected to increase four times by 2040 (Teskey et al., 2015). Rising temperatures pose a substantial threat to urban tree health, disrupting key physiological processes such as photosynthesis. Heat stress reduces photosynthetic efficiency, slowing down carbon assimilation and limiting tree growth and survival (Ahrens et al., 2021; Sharkey, 2005). Research shows that by 2099, 82 out of 140 urban tree species currently thriving in California would be rendered unfit for planting due to the region's expected temperature increases (McBride and Laćan 2018). Similarly, studies in Europe have shown that certain tree species, such as *Fagus sylvatica*, are already experiencing stunted growth in southern regions due to rising temperatures (Schulze et al., 2005). In the United Kingdom, heatwaves in recent years have resulted in thousands of excess deaths, widespread damage to infrastructure and forests, with southern regions facing particularly harsh conditions (Kendon et al., 2021, 2022).

The effects of elevated temperatures on trees at the leaf scale can be assessed with parameters derived from the light response curve through chlorophyll fluorescence methods (Benedetti et al. 2018; Pérez-Bueno et al. 2019). These parameters provide valuable insights into how efficiently plants convert absorbed light into energy and dissipate excess energy under stress. Key indicators include non-photochemical quenching (NPQ), a process in which excess absorbed light energy is dissipated as heat to protect plants from photodamage (Li

et al., 2015; Ruban, 2016), and Electron transport rate (ETR_{max}), which indicates photosynthetic capacity and efficiency, with higher values showing better stress resilience (Baker, 2008; Shanker et al., 2022). Chlorophyll fluorescence analysis is widely used in research areas such as plant breeding, stress physiology, and ecosystem monitoring. With the increasing need to optimise crop yields and improve plant resilience to climate change, understanding photosynthesis on a deeper level has become more important than ever (Cochavi et al., 2020).

A growing body of research suggests that trees exhibit delayed physiological responses to heat stress, where recovery extends well beyond the actual heat event (Rijnhart et al., 2022). These temporal dynamics range from immediate responses like stomatal conductance changes to longer-term adjustments in photosynthetic capacity, also known as time lag effects, taking days or weeks (Sage & Kubien, 2007). (Zhu et al., 2023) demonstrated that response lag times vary by specific physiological process, while showing that these responses interact with plants' circadian rhythms. Understanding these time-lagged effects is essential for predicting tree performance under increasingly variable climate conditions, particularly in urban environments where heat stress events are becoming more frequent and intense.

Despite increasing research on heat stress in trees, key gaps remain in understanding how these delayed responses manifest in urban settings. While previous studies have examined tree responses to heat across various species and contexts ((Feeley et al., 2020; Flexas et al., 2004; Jiang & Huang, 2000; Krause et al., 2010; Li et al., 2015; Nankishore & Farrell, 2016; Oliveira & Peñuelas, 2005; Pociecha et al., 2008; Sage & Kubien, 2007; Teskey et al., 2015; Xu et al., 2007; Zhang et al., 2014; Zhu et al., 2023) much of the existing work has been conducted in natural forests or agricultural settings, with limited studies examining the physiological resilience of trees specifically in urban microclimates. Moreover, the majority of these studies do not consider the lagged response between variables.

To address these gaps, our study investigates the time-lagged effects of heat stress on a common urban tree in the UK, *Acer campestre* or field maple, with a focus on physiological parameters such as NPQ and ETR_{max}. Through controlled mesocosm experiments, we systematically measured changes in these indicators through summer seasons of 2022 and 2023, quantifying the duration and severity of delayed stress responses. By identifying prolonged physiological impairments, we highlight the potential for cumulative heat stress impacts, which may be more severe than previously understood. These findings have critical implications for urban tree selection, climate adaptation strategies, and the design of resilient urban green spaces in the face of escalating heat extremes.

The field maple, belonging to the Sapindaceae family, is a native species of Britain and much of continental Europe. It typically grows up to 15 metres tall (occasionally reaching 25 m) and can live for several decades. Commonly found in hedgerows, woodlands, parklands, and wood pastures across the UK, it has light grey bark that flakes with age and forms a low, rounded crown. Its bright green leaves turn golden or red in autumn. Yellow-green flowers appear in late April, followed by winged fruits (samaras) that ripen in September and disperse in October. Seeds take about 18 months to germinate, with rapid growth occurring between 5 and 25 years. Pollination is mainly insect-driven, though some occurs via wind (Zecchin et al., 2016).

Methodology

Our study is divided into two phases. In the 2022 laboratory phase, the controlled conditions allowed us to establish baseline measurements for key traits and temperature responses without environmental interference. This was crucial for isolating variables and understanding fundamental relationships. The transition to field measurements in 2023 added ecological validity by capturing how these physiological processes operate under natural conditions, including daily and seasonal fluctuations. This approach bridged controlled experimentation with real-world application.

Tree species of study

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We purchased the field maple trees in this study from Barcham Trees, Nursery in Soham, Cambridgeshire, England. They specialise in container trees and has been providing their services since thirty-five years. In 2024, they were awarded royal warrant as specialist container tree growers by King Charles III of U.

Setup for the experimental trees

Eight field maple trees were placed in an array of 90-litre pots connected to an irrigation system based on the experiment by Araya et al (2010). The system was designed to maintain a constant water table and ensure trees were not water-limited. Each pot had layers of gravel, sand, and soil, with each layer separated by a weed membrane (Fig. 1a). The pots were set at a consistent level using a laser level (Fig. 1b). This setup allowed for a clearer understanding of temperature stress responses independent of water availability.

The trees were tied to a metal cable to keep them upright, with hoses connecting them to a small water chamber that maintained the level of water in the pots via capillary action, which in turn was connected to a large water reservoir for a constant water supply (Fig. 1d).

Data collection protocol for 2022

Measurements were conducted on days with dry weather at 10:00 am and 04:00 PM using the OSp5 + modulated chlorophyll fluorometer (Opti-Sciences, 2013). Leaves were collected using a pole lopper, with stems cut underwater to prevent air embolism, and maintained in water-filled centrifuge tubes during testing. For each leaf, dark-adapted fluorescence values were recorded first as baseline, followed by rapid light curve measurements at PAR levels of 10–180 µmol m^2s^1, with three-minute intervals between readings (Haldimann & Feller, 2004), shown in Fig. 2.

Data collection protocol for 2023

Weekly measurements required a tele-tower assembly to access the tree canopy (Fig. 1c), with setup assistance from lab staff and preparation beginning one day before data collection. Dark-adaptation clips were placed on target leaves 24 hours before measurements. Data was collected at three timepoints (09:30 am, 1:00 pm, and 4:30 pm), beginning with dark-adapted fluorescence readings followed by yield measurements from the same spot on leaves throughout the day.

Statistical analysis

Parameter extraction

The lm package in R (Elzhov et al., 2023) was used to fit the non-rectangular hyperbola equation (Eq. 1) to PAR vs ETR data and extract the required parameters (Ye et al., 2013).

$$ETR = \frac{\alpha * PAR + ETRmax - \sqrt{(\alpha * PAR + ETRmax)^2 - 4 * \theta * \alpha * PAR * ETRmax}}{2 * \theta} \quad \text{Eq. 1}$$

The α in Eq. 1 represents the initial slope of the curve, which symbolises the photosynthetic efficiency at low light levels. ETR_{max} represents the saturation point in plant photosynthesis. θ is the factor of curvature that accounts for the transition from linear to saturation.

$$NPQ = \frac{Fm - Fm'}{Fm'} \quad \text{Eq. 2}$$

Fm in Eq. 2 is the maximum fluorescence yield in the dark-adapted state, and Fm' is the maximum fluorescence yield in the light-adapted state (Opti-Sciences, 2013).

Modelling the data

The 2022 data had repeated measurements and seasonality. Generalised linear mixed models (GLMMs) are statistical models that account for the non-independence of errors among observations that arise due to repeated measures and clustering. The lme4 package in R was used for fitting the generalized linear mixed models to the time series data (Bolker et al., 2009; D. Wang et al., 2016).

The CCF function in R (Brockwell & Davis, 1991) was used to analyse the lag between leaf temperature and corresponding extracted parameters.

The 2023 data was sparse, hence a non-parametric statistical method called Spearman's rank correlation was used to ascertain the direction and strength of the relationship between two variables. This method is appropriate for data with seasonal trends and a small sample size since it is less susceptible to outliers and does not presuppose a linear relationship between the variables. In R, the base package ‘stats’ is used for this correlation method.

Results

Season 2022

Measurements from the 2022 season encompassed 240 rapid light curves (RLCs) derived from eight leaves per day across 30 days. From these, we extracted ETR_{max}, Alpha, and theta values, while NPQ values were calculated using dark-adapted measurements taken before actinic light exposure using Eq. 2.

Time series analysis revealed a distinct temporal relationship between NPQ and leaf temperature (Fig. 3). To quantify this relationship, we performed cross-correlation function (CCF) analysis on morning and evening data separately.

Morning measurements exhibited significant lag effects at 0, -1, and − 2 days (Fig. 4a), suggesting that NPQ response preceded leaf temperature change. Evening data showed no significant lag effect (Fig. 4b), indicating synchronous responses between variables during this period.

To further investigate this time-dependent relationship, we implemented a generalised mixed-effect model incorporating the observed lag effects for morning data (Table 1). Results revealed that same-day leaf temperature (lag 0) had a statistically significant negative relationship with NPQ ($\beta = -0.03$, $p = 0.02$). Specifically, for each unit increase in leaf temperature, NPQ decreased by approximately 0.03 units. Despite the presence of lag effects at -1 and − 2 days in CCF analysis, these lagged temperature effects did not demonstrate statistical significance in the model (lag 1: $\beta = -0.00001$, $p = 0.91$; lag 2: $\beta = 0.002$, $p = 0.80$).

Table 1
Influence of lagged temperature effects on NPQ in morning measurements (2022)

Predictors	Estimates	std. Error	p-value
(Intercept)	1.18	0.39	**0.007**
lag 0	-0.03	0.01	**0.02**
lag 1	-0.00001	0.01	0.91
lag 2	0.002	0.01	0.80

Similar temporal dynamics were observed between ETR_{max} and leaf temperature (Fig. 5), but with different lag patterns. Morning data revealed a significant lag effect at -2 days (Fig. 6a), indicating that leaf temperature changes preceded ETR_{max} responses by approximately two days. Generalised linear mixed effect modelling of this relationship demonstrated that for each unit increase in leaf temperature (at lag − 2), ETR_{max} decreased by 0.08 units ($p = 0.046$).

In contrast, evening measurements showed no significant lag between ETR_{max} and leaf temperature (Fig. 6b), consistent with the pattern observed for NPQ. This suggests a fundamental difference in the temporal dynamics of photosynthetic responses between morning and evening conditions.

Season 2023

Light response curves were fitted using the non-rectangular hyperbola equation (Eq. 1) to the entire season data. Parameters extracted from these curves showed distinct diurnal patterns (Table 2; Fig. 7).

Table 2
Physiological parameters at different times of day in 2023

	Time of Day	09:30 am	01:00 pm	04:30 pm
Alpha	Value	0.19	0.21	0.19
	t_values	3.18	3.84	22.67
	p_values	**0.002**	**< 0.001**	**< 0.001**
ETR_{max}	Value	14.84	18.97	21.92
	t_values	6.80	7.33	19.06
	p_values	**< 0.001**	**< 0.001**	**< 0.001**
Theta	Value	0.74	0.87	1.004
	t_values	1.85	3.87	36.39
	p_values	0.06	**< 0.001**	**< 0.001**

The initial quantum yield (Alpha) exhibited a midday peak of 0.2144 at 01:00 pm compared to morning and late afternoon values of 0.19 at 09:30 am and 04:30 pm. This suggested enhanced light utilisation efficiency at midday. All Alpha values were statistically significant ($p < 0.05$), with the strongest significance observed at 04:30 pm ($t = 22.67$, $p < 0.001$).

Maximum electron transport rate (ETR_{max}) demonstrated a progressive increase throughout the day, from 14.84 at 09:30 am to 18.97 at 01:00 pm, reaching a maximum of 21.92 at 04:30 pm. This pattern indicated intensifying photosynthetic capacity as the day advanced. All ETR_{max} values showed high statistical significance ($p < 0.001$), with the highest confidence at 04:30 pm ($t = 19.06$).

The convexity parameter (theta) followed an increasing trend from morning (0.74) through midday (0.87) to late afternoon (1.004), suggesting progressively greater light saturation as the day progressed. While midday and afternoon theta values were highly significant ($p < 0.001$), the morning value did not reach statistical significance ($t = 1.85$, $p = 0.06$).

Time series analysis of NPQ throughout the 2023 season revealed elevated values in early July (Fig. 8), indicating increased thermal energy dissipation during this period. However, examination of the relationship between NPQ and leaf temperature at distinct diurnal time points (09:30 am, 01:00 pm, and 04:30 pm) revealed no statistically significant correlations (Table 3).

Table 3
Spearman correlation between NPQ and leaf temperature at separate times of day

Time of Day	p values	Spearman's correlation
09.30 am	0.31	0.14
01.00 pm	0.84	0.03
04.30 pm	0.88	0.02

Discussion

Our study examined the temporal dynamics of heat stress responses in *Acer campestre*, focusing on chlorophyll fluorescence parameters across two contrasting summer seasons and different times of day. By analysing both short-term lag effects and diurnal variation, we inspected how photosynthetic function in this common urban tree was shaped not only by ambient temperature but also by its recent thermal history.

In 2022, under controlled mesocosm conditions and in a year marked by record-breaking heat in the UK (Dessai et al., 2024), our findings demonstrated significant time-lagged responses in key physiological parameters, particularly during morning measurements. NPQ displayed statistically significant lags at 0, -1, and − 2 days, with a same-day negative association with leaf temperature (β = -0.03, $p = 0.026$), suggesting reduced photoprotective capacity under thermal stress. ETRmax exhibited an anticipatory decline two days before peak leaf temperature (β = -0.08, $p = 0.046$), indicating a pre-emptive downregulation of photosynthesis potentially to minimise photodamage during periods of rising heat. These results suggested that *Acer campestre* engages in proactive and delayed physiological adjustment as a protective mechanism in response to cumulative heat exposure.

The lag effects were absent in evening measurements. This contrast pointed to a diurnal divergence in physiological behaviour, where the morning period may represent a critical window for heat response modulation. The absence of lag in the evening may indicate physiological equilibration after sustained exposure, or that protective mechanisms are depleted or have adapted to the thermal load accumulated during the day.

In 2023, when ambient conditions were notably milder, the physiological patterns diverged significantly. ETRmax values increased progressively across the day, from 14.84 in the morning to 21.92 in the late afternoon ($p < 0.001$), suggesting enhanced photosynthetic capacity in the absence of extreme thermal constraints. NPQ values peaked during early July, yet no significant correlations were found between NPQ and leaf temperature across all three time points. These findings imply that in the absence of thermal extremes, *Acer campestre* maintains stable diurnal physiological rhythms without the disruption or delayed responses observed in the previous year.

The contrasting responses observed between 2022, a year of record-breaking temperatures, and 2023, which experienced more moderate conditions, highlight the importance of studying tree physiology across different thermal regimes. This comparative approach helps identify physiological thresholds beyond which adaptive mechanisms may fail, offering critical insights for species selection and urban forest management.

As climate change intensifies, urban and natural forests face increasing challenges from rising temperatures and extreme weather events. Urban trees, in particular, are highly vulnerable to extreme heat, which can accelerate senescence and increase mortality rates (Moser-Reischl et al., 2018). The ability of trees to tolerate thermal stress depends on the frequency, intensity, and duration of high-temperature events, making it essential to assess their physiological limits (Ahrens et al., 2021). Research suggests that prioritising species diversity and high-quality selections over simply increasing tree numbers can enhance resilience to climate stressors (Stevenson et al., 2020).

Our findings underscore the importance of heat intensity and time of day in shaping physiological outcomes. The consistent morning-specific lag effects, especially under severe heat conditions, suggest the presence of a form of “thermal memory” in *Acer campestre*, where past environmental conditions influence present-day function. This memory may involve slow-recovering biochemical changes, altered energy allocation, or stress priming processes, which enable the tree to adjust its photoprotective mechanisms not in real time but in anticipation of, or response to, previously experienced conditions.

From an applied perspective, these findings hold practical relevance for urban forestry. Trees planted in dense urban environments face compounded stressors, including the urban heat island effect, limited rooting space, and inconsistent watering (Chaston et al., 2022; Tan et al., 2016; X. Wang et al., 2021; Y. Wang & Akbari, 2016). Understanding that heat responses are not always immediate and may differ markedly between morning and evening suggests that physiological assessments and management interventions should be timed accordingly. Morning appears to be a particularly sensitive period, where trees exhibit both anticipatory and delayed adjustments, and where targeted support may be most beneficial.

Despite the novel insights, this study is not without limitations. First, the sample size, while appropriate for physiological measurements, limits broader generalisation. Data from 2023, collected in natural conditions, were sparser and less evenly distributed than the controlled 2022 dataset, potentially influencing the detection of lag effects. Additionally, this study focuses solely on leaf-level fluorescence responses. While these are critical indicators of stress, they provide only part of the picture. Other physiological processes, such as stomatal conductance, water potential, or carbohydrate allocation, may interact with or buffer the effects we observed and were not assessed in this study. Furthermore, the observed patterns are specific to one species, *Acer campestre*. Other urban tree species may respond differently in terms of both lag magnitude and diurnal sensitivity.

Future studies should build on these findings by incorporating a broader range of tree species to determine whether the observed lag effects are specific to *Acer campestre* or reflect more general physiological strategies among urban trees. Expanding the temporal scale of data collection across multiple growing seasons, particularly those featuring varied heat stress profiles, would help to capture interannual variability and better understand the cumulative impact of repeated or compound heatwave events. Additionally, integrating whole-tree and ecosystem-level measurements such as canopy temperature, transpiration rates, and growth performance could link leaf-level responses to longer-term indicators of tree health and resilience. To fully elucidate the mechanisms underpinning observed lag effects and the proposed concept of “thermal memory,” future research should also explore molecular and biochemical pathways, including the roles of heat shock proteins, antioxidant systems, and potential epigenetic modifications that may mediate delayed physiological responses under thermal stress.

Conclusion

This study reveals key temporal patterns in *Acer campestre's* response to heat stress, with distinct differences between morning and evening physiological responses and across two contrasting summer seasons. Using chlorophyll fluorescence techniques, we identified significant lag effects between leaf temperature and key photosynthetic parameters, specifically in the 2022 mesocosm phase, when extreme heat conditions prevailed.

Morning measurements showed that NPQ responded with notable lags at 0, -1, and − 2 days, with a significant same-day negative correlation with leaf temperature (β = -0.03, $p = 0.026$), suggesting a diminished capacity for thermal energy dissipation as temperature rises. ETRmax also exhibited a significant two-day anticipatory lag (β = -0.08, $p = 0.046$), implying that heat stress effects on photosynthesis begin before temperature peaks. These results were absent in evening data, indicating that trees exhibit more synchronised, real-time physiological responses later in the day.

In 2023, field-based measurements captured diurnal changes in photosynthetic activity without evidence of lag. ETRmax increased steadily from morning to afternoon (from 14.84 to 21.92, $p < 0.001$), and NPQ values peaked in early July but were not significantly linked to temperature at any time point. This contrast between years emphasises the role of heat intensity in disrupting physiological rhythms and the importance of monitoring time-of-day dynamics.

The consistent observation of morning-specific lags suggests the presence of a short-term "thermal memory," where past temperature conditions influence current-day physiological performance. These insights have direct relevance for urban tree management, highlighting morning as a particularly sensitive window and suggesting that adaptive strategies such as irrigation timing and species placement should be informed by diurnal physiological profiles.

Future work should explore whether these lagged patterns are consistent across species and climatic conditions and assess how long-term cumulative effects shape urban tree resilience in an era of intensifying heat stress.

Abbreviations

Sustainable Development Goals (SDGs), Non-Photochemical Quenching (NPQ), Electron Transport Rate (ETR), Generalised Linear Mixed Model (GLMM), Rapid Light Curve (RLC), Photosynthetic Aperture Radar (PAR), Cross Correlation Function (CCF).

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Data Availability: The data that support the findings of this study are openly available in Ramla101/Plant-Physiology-data-for-2022-and-2023 at https://zenodo.org/records/15051050.

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References

1. Ahrens CW, Challis A, Byrne M, Leigh A, Nicotra AB, Tissue D, Rymer P. Repeated extreme heatwaves result in higher leaf thermal tolerances and greater safety margins. New Phytol. 2021;232(3):1212–25. https://doi.org/10.1111/nph.17640.
2. Araya YN, Gowing DJ, Dise N. A controlled water-table depth system to study the influence of fine-scale differences in water regime for plant growth. Aquat Bot. 2010;92(1):70–4. https://doi.org/10.1016/j.aquabot.2009.10.004.
3. Armson D, Stringer P, Ennos AR. (2012). The Effect of Tree Shade and Grass on Surface and Globe Temperatures in An Urban Area. In *URBAN FORESTRY & URBAN GREENING, 2012*.
4. Baker NR. Chlorophyll fluorescence: A probe of photosynthesis in vivo. Annu Rev Plant Biol. 2008;59:89–113. https://doi.org/10.1146/annurev.arplant.59.032607.092759.
5. Benedetti M, Vecchi V, Barera S, Dall'Osto L. (2018). Biomass from microalgae: The potential of domestication towards sustainable biofactories. *Microbial Cell Factories*, *17*. https://doi.org/10.1186/s12934-018-1019-3
6. Bolker BM, Brooks ME, Clark CJ, Geange SW, Poulsen JR, Stevens MHH, White JS S. Generalized linear mixed models: A practical guide for ecology and evolution. Trends Ecol Evol. 2009;24(3):127–35. https://doi.org/10.1016/j.tree.2008.10.008.
7. Brockwell PJ, Davis RA. Time Series: Theory and Methods: Theory and Methods. Springer Science & Business Media; 1991.
8. Chaston T, Broome R, Cooper N, Duck G, Geromboux C, Yuming Guo FJ, Perkins-Kirkpatrick S, Zhang Y, Morgan GSD, G., Hanigan I. Mortality Burden of Heatwaves in Sydney, Australia Is Exacerbated By The Urban Heat Island and Climate. Can Tree Cover Help Mitigate The Health Impacts? ATMOSPHERE; 2022.
9. Cochavi A, Amer M, Stern R, Yakir D. (2020). Using Sun-induced fluorescence and Carbonyl Sulfide flux to assess the response to seasonal heatwave in a citrus orchard. *22nd EGU General Assembly, Held Online 4–8 May, 2020, Id.8440*. https://doi.org/10.5194/egusphere-egu2020-8440
10. Coutts AM, White EC, Tapper NJ, Beringer J, Livesley SJ. Temperature and Human Thermal Comfort Effects of Street Trees Across Three Contrasting Street Canyon Environments. THEORETICAL AND APPLIED CLIMATOLOGY; 2015.
11. Dessai S, Lonsdale K, Lowe J, Harcourt R, editors. Quantifying Climate Risk and Building Resilience in the UK. Springer International Publishing; 2024. https://doi.org/10.1007/978-3-031-39729-5.

12. Elzhov TV, Mullen KM, Spiess A-N, Bolker B. (2023). *R Interface to the Levenberg-Marquardt Nonlinear Least-Squares Algorithm Found in MINPACK, Plus Support for Bounds* (p. 1.2-4) [Dataset]. https://doi.org/10.32614/CRAN.package.minpack.lm
13. Feeley K, Martinez-Villa J, Perez T, Silva Duque A, Triviño Gonzalez D, Duque A. The Thermal Tolerances, Distributions, and Performances of Tropical Montane Tree Species. Front Forests Global Change. 2020;3. https://doi.org/10.3389/ffgc.2020.00025.
14. Flexas J, Bota J, Loreto F, Cornic G, Sharkey TD. Diffusive and metabolic limitations to photosynthesis under drought and salinity in C3 plants. Plant Biol. 2004;6(3):269–79. https://doi.org/10.1055/s-2004-820867.
15. Haldimann P, Feller U. Inhibition of photosynthesis by high temperature in oak (Quercus pubescens L.) leaves grown under natural conditions closely correlates with a reversible heat-dependent reduction of the activation state of ribulose-1,5-bisphosphate carboxylase/oxygenase. Plant Cell Environ. 2004;27(9):1169–83. https://doi.org/10.1111/j.1365-3040.2004.01222.x.
16. IPCC. In: Zhai, editor. Synthesis report of the IPCC sixth assessment report (AR6). D. K. D. P.; 2023.
17. Jiang Y, Huang B. (2000). Effects of Drought Or Heat Stress Alone and in Combination on Kentucky Bluegrass. Crop Sci. https://www.paperdigest.org/paper/?paper_id=doi.org_10.2135_cropsci2000.4051358x
18. Kaplan R. Impact of Urban Nature: A Theoretical Analysis. URBAN ECOLOGY; 1984.
19. Kendon M, McCarthy M, Jevrejeva S, Matthews A, Sparks T, Garforth J. State of the UK Climate 2020. Int J Climatol. 2021;41(S2):1–76. https://doi.org/10.1002/joc.7285.
20. Kendon M, McCarthy M, Jevrejeva S, Matthews A, Sparks T, Garforth J, Kennedy J. State of the UK Climate 2021. Int J Climatol. 2022;42(S1):1–80. https://doi.org/10.1002/joc.7787.
21. Krause GH, Winter K, Krause B, Jahns P, García M, Aranda J, Virgo A. High-temperature tolerance of a tropical tree, Ficus insipida: Methodological reassessment and climate change considerations. Funct Plant Biol. 2010;37(9):890–900. https://doi.org/10.1071/FP10034.
22. Li S, Yang W, Yang T, Chen Y, Ni W. (2015). Effects of Cadmium Stress on Leaf Chlorophyll Fluorescence and Photosynthesis of Elsholtzia Argyi—A Cadmium Accumulating Plant. *INTERNATIONAL JOURNAL OF PHYTOREMEDIATION*. https://www.paperdigest.org/paper/?paper_id=doi.org_10.1080_15226514.2013.828020
23. McBride JR, Laćan I. The impact of climate-change induced temperature increases on the suitability of street tree species in California (USA) cities. Urban Forestry Urban Green. 2018;34:348–56. https://doi.org/10.1016/j.ufug.2018.07.020.
24. Meier F, Scherer D. Spatial and Temporal Variability of Urban Tree Canopy Temperature During Summer 2010 in. THEORETICAL AND APPLIED CLIMATOLOGY; 2012.
25. Moser-Reischl A, Uhl E, Rötzer T, Biber P, Con T, Tan NT, Pretzsch H. Effects of the urban heat Island and climate change on the growth of Khaya senegalensis in Hanoi. Vietnam For Ecosyst. 2018;5(1):1–14. https://doi.org/10.1186/S40663-018-0155-X/TABLES/7.
26. Nankishore A, Farrell AD. (2016). The Response Of Contrasting Tomato Genotypes To Combined Heat And Drought Stress. *JOURNAL OF PLANT PHYSIOLOGY*. https://www.paperdigest.org/paper/?paper_id=pubmed-27467552

27. Oliveira G, Peñuelas J. (2005). Effects of Winter Cold Stress on Photosynthesis and Photochemical Efficiency of PSII of The Mediterranean Cistus Albidus L. and Quercus Ilex L. *PLANT ECOLOGY*. https://www.paperdigest.org/paper/?paper_id=doi.org_10.1007_s11258-005-4876-x
28. Opti-Sciences. (2013). *OS5p + User's Guide*. www.optsci.com.
29. Pociecha E, Kościelniak J, Filek W. (2008). Effects of Root Flooding and Stage of Development on The Growth and Photosynthesis of Field Bean (Vicia Faba L. Minor). *ACTA PHYSIOLOGIAE PLANTARUM*. https://www.paperdigest.org/paper/?paper_id=doi.org_10.1007_s11738-008-0151-9
30. Rijnhart JJM, Twisk JWR, Valente MJ, Heymans MW. Time lags and time interactions in mixed effects models impacted longitudinal mediation effect estimates. J Clin Epidemiol. 2022;151:143–50. https://doi.org/10.1016/j.jclinepi.2022.07.004.
31. Ruban AV. Nonphotochemical Chlorophyll Fluorescence Quenching: Mechanism and Effectiveness in Protecting Plants from Photodamage1. Plant Physiol. 2016;170(4):1903–16. https://doi.org/10.1104/pp.15.01935.
32. Sage RF, Kubien DS. The temperature response of C3 and C4 photosynthesis. Plant Cell Environ. 2007;30(9):1086–106. https://doi.org/10.1111/J.1365-3040.2007.01682.X.
33. Schulze E-D, Beck E, Müller-Hohenstein K. Plant Ecology. Springer Science & Business Media; 2005.
34. Shanker AK, Amirineni S, Bhanu D, Yadav SK, Jyothilakshmi N, Vanaja M, Singh J, Sarkar B, Maheswari M, Singh VK. (2022). High-resolution dissection of photosystem II electron transport reveals differential response to water deficit and heat stress in isolation and combination in pearl millet [Pennisetum glaucum (L.) R. Br.]. *Frontiers in Plant Science*, *13*. https://doi.org/10.3389/fpls.2022.892676
35. Sharkey TD. Effects of moderate heat stress on photosynthesis: Importance of thylakoid reactions, rubisco deactivation, reactive oxygen species, and thermotolerance provided by isoprene. Plant Cell Environ. 2005;28(3):269–77. https://doi.org/10.1111/j.1365-3040.2005.01324.x.
36. Stevenson PC, Bidartondo MI, Blackhall-Miles R, Cavagnaro TR, Cooper A, Geslin B, Koch H, Lee MA, Moat J, O'Hanlon R, Sjöman H, Sofo A, Stara K, Suz LM. (2020). The state of the world's urban ecosystems: What can we learn from trees, fungi, and bees? *PLANTS, PEOPLE, PLANET*, *2*(5), 482–498. https://doi.org/10.1002/ppp3.10143
37. Tan Z, Lau KK-L, Ng E. Urban Tree Design Approaches for Mitigating Daytime Urban Heat Island Effects in A High-density Urban Environment. ENERGY AND BUILDINGS; 2016.
38. Teskey R, Wertin T, Bauweraerts I, Ameye M, Mcguire MA, Steppe K. Responses of tree species to heat waves and extreme heat events. Plant Cell Environ. 2015;38(9):1699–712. https://doi.org/10.1111/pce.12417.
39. Turner-Skoff JB, Cavender N. The benefits of trees for livable and sustainable communities. PLANTS PEOPLE PLANET. 2019;1(4):323–35. https://doi.org/10.1002/ppp3.39.
40. Wang D, Heckathorn SA, Mainali K, Tripathee R. Timing Effects of Heat-Stress on Plant Ecophysiological Characteristics and Growth. Front Plant Sci. 2016;7:1629. https://doi.org/10.3389/fpls.2016.01629.
41. Wang X, Dallimer M, Scott CE, Shi W, Gao J. Tree Species Richness and Diversity Predicts The Magnitude of Urban Heat Island Mitigation Effects of Greenspaces. THE SCIENCE OF THE TOTAL ENVIRONMENT; 2021.

42. Wang Y, Akbari H. The Effects of Street Tree Planting on Urban Heat Island Mitigation in Montreal. SUSTAINABLE CITIES AND SOCIETY; 2016.
43. Xu ZZ, Zhou GS, Wang YL, Han GX, Li YJ. (2007). Changes in Chlorophyll Fluorescence in Maize Plants with Imposed Rapid Dehydration at Different Leaf Ages. *JOURNAL OF PLANT GROWTH REGULATION*. https://www.paperdigest.org/paper/?paper_id=doi.org_10.1007_s00344-007-9035-2
44. Ye Z-P, Suggett DJ, Robakowski P, Kang H-J. A mechanistic model for the photosynthesis–light response based on the photosynthetic electron transport of photosystem II in C3 and C4 species. New Phytol. 2013;199(1):110–20. https://doi.org/10.1111/nph.12242.
45. Zecchin B, Caudullo G, de Rigo D. (2016). *Acer campestre in Europe: Distribution, habitat, usage and threats*.
46. Zhang Y-J, Holbrook NM, Cao K-F. (2014). Seasonal Dynamics In Photosynthesis Of Woody Plants At The Northern Limit Of Asian Tropics: Potential Role Of Fog In Maintaining Tropical Rainforests And Agriculture In Southwest China. Tree Physiol. https://www.paperdigest.org/paper/?paper_id=pubmed-25298374
47. Zhu L, Scafaro AP, Vierling E, Ball MC, Posch BC, Stock F, Atkin OK. (2023). Heat tolerance of a tropical-subtropical rainforest tree species Polyscias elegans: Time-dependent dynamic responses of physiological thermostability and biochemistry. *The New Phytologist*. https://api.semanticscholar.org/CorpusID:265041915.

Figures

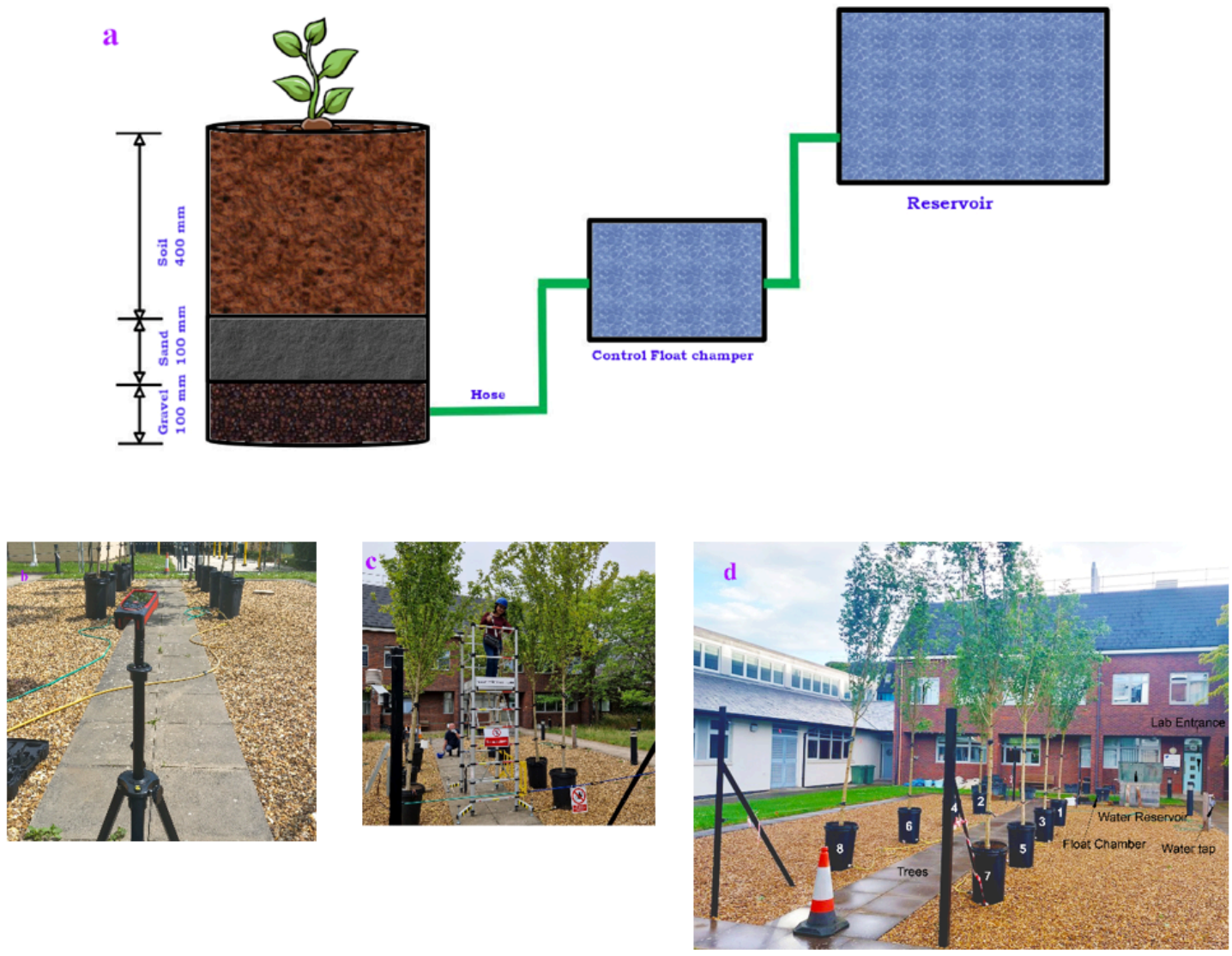

Figure 1

Experimental setup; a) Controlled water depth system, b) Laser leveller, c) Researchers using a tele-tower for data collection, d) numbered trees with labelled setup.

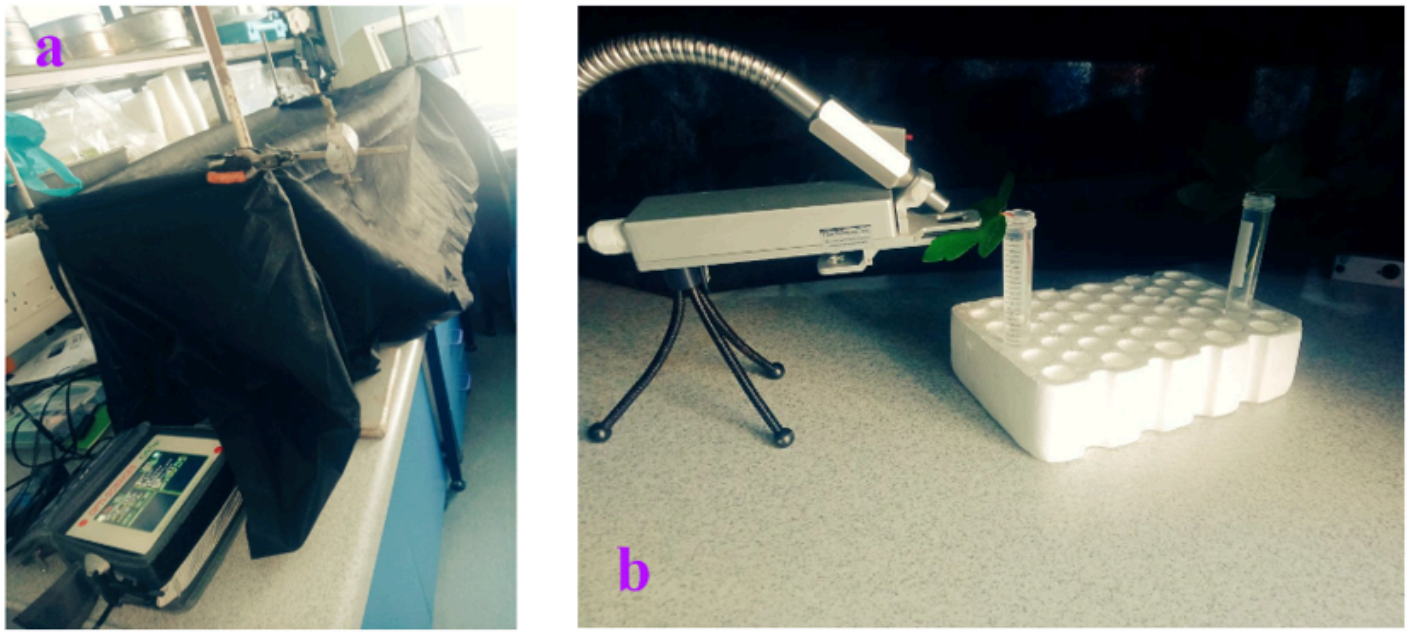

Figure 2

Dark-adapted setup for collecting 2022 fluorescence data using OSp5+ fluorometer

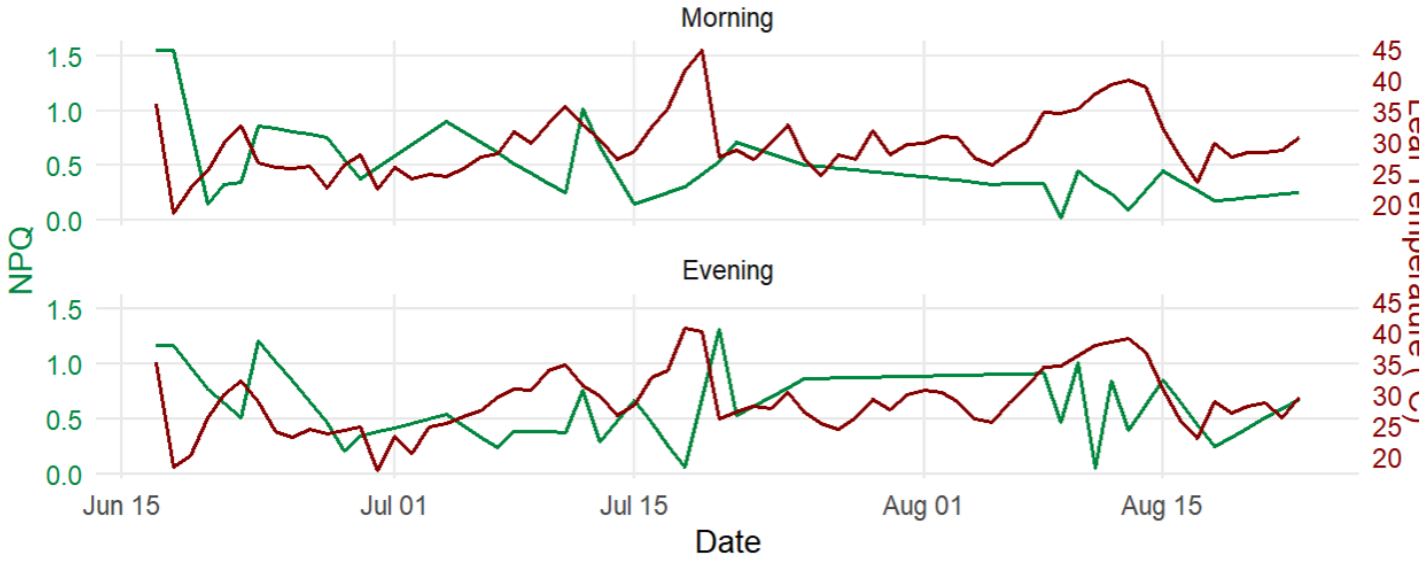

Figure 3

NPQ and Leaf temperature values in 2022 during morning and evening time

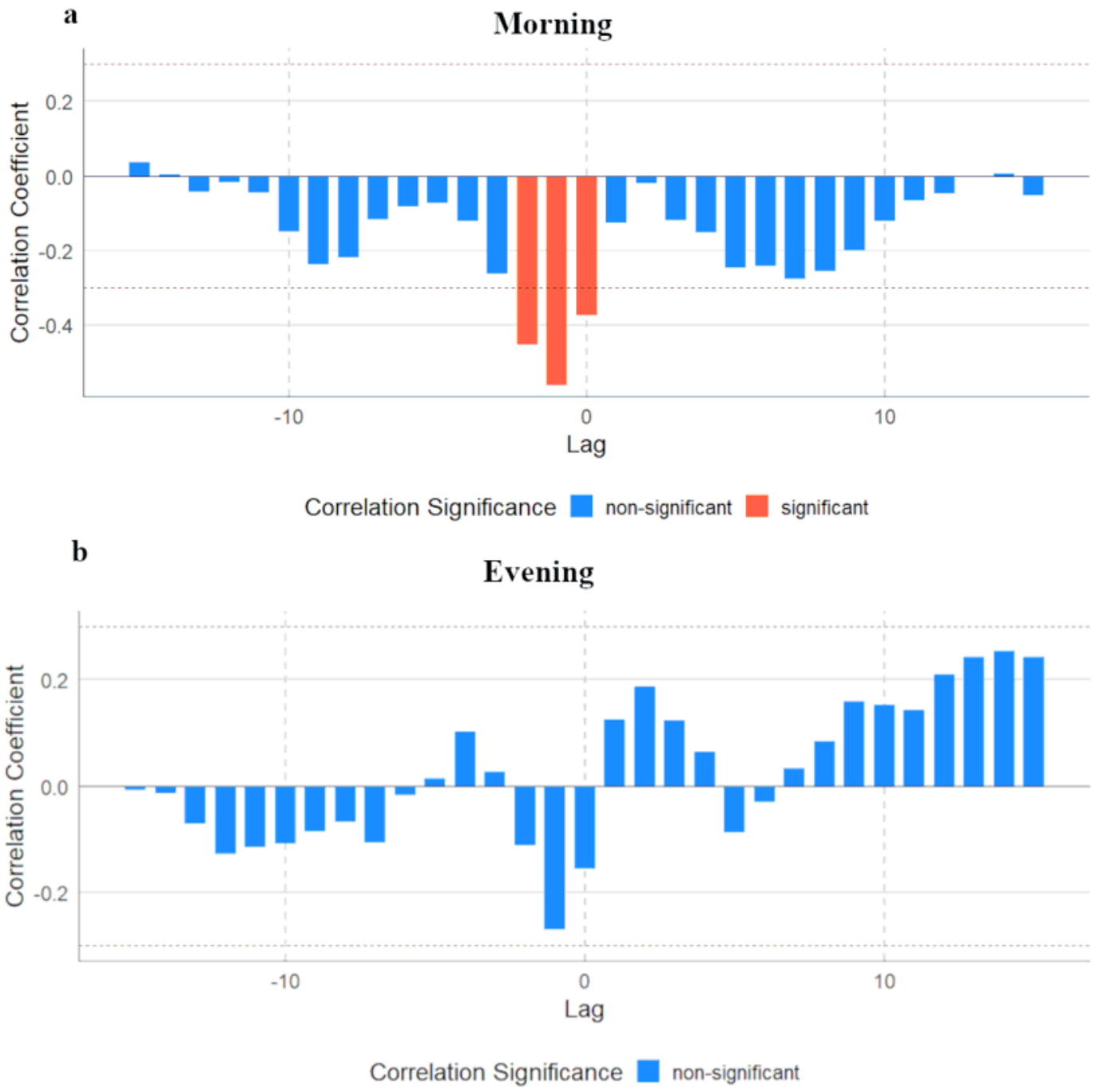

Figure 4

Lag effect between NPQ and leaf temperature in 2022: a) Morning, b) Evening

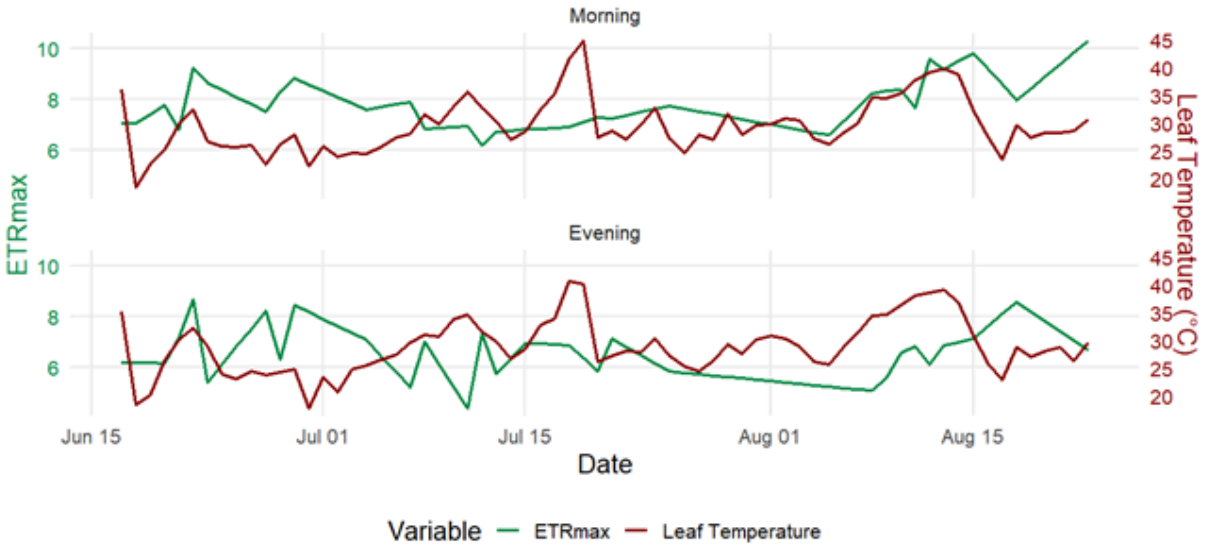

Figure 5

TS plot ETR_{max} and Leaf temperature values in 2022 during morning and evening time

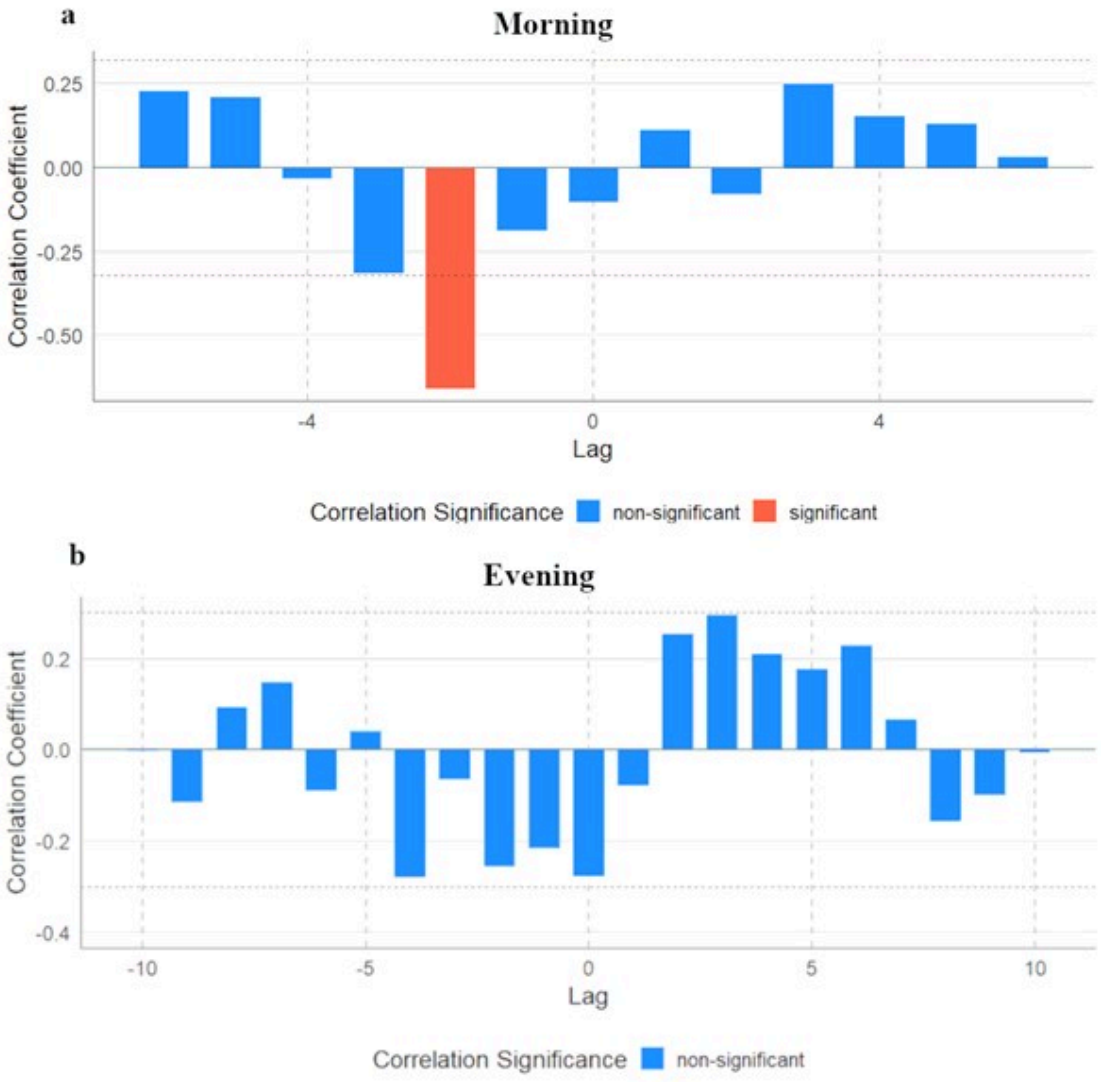

Figure 6

Lag effect between ETR_{max} and Leaf temperature in 2022: a) Morning, b) Evening

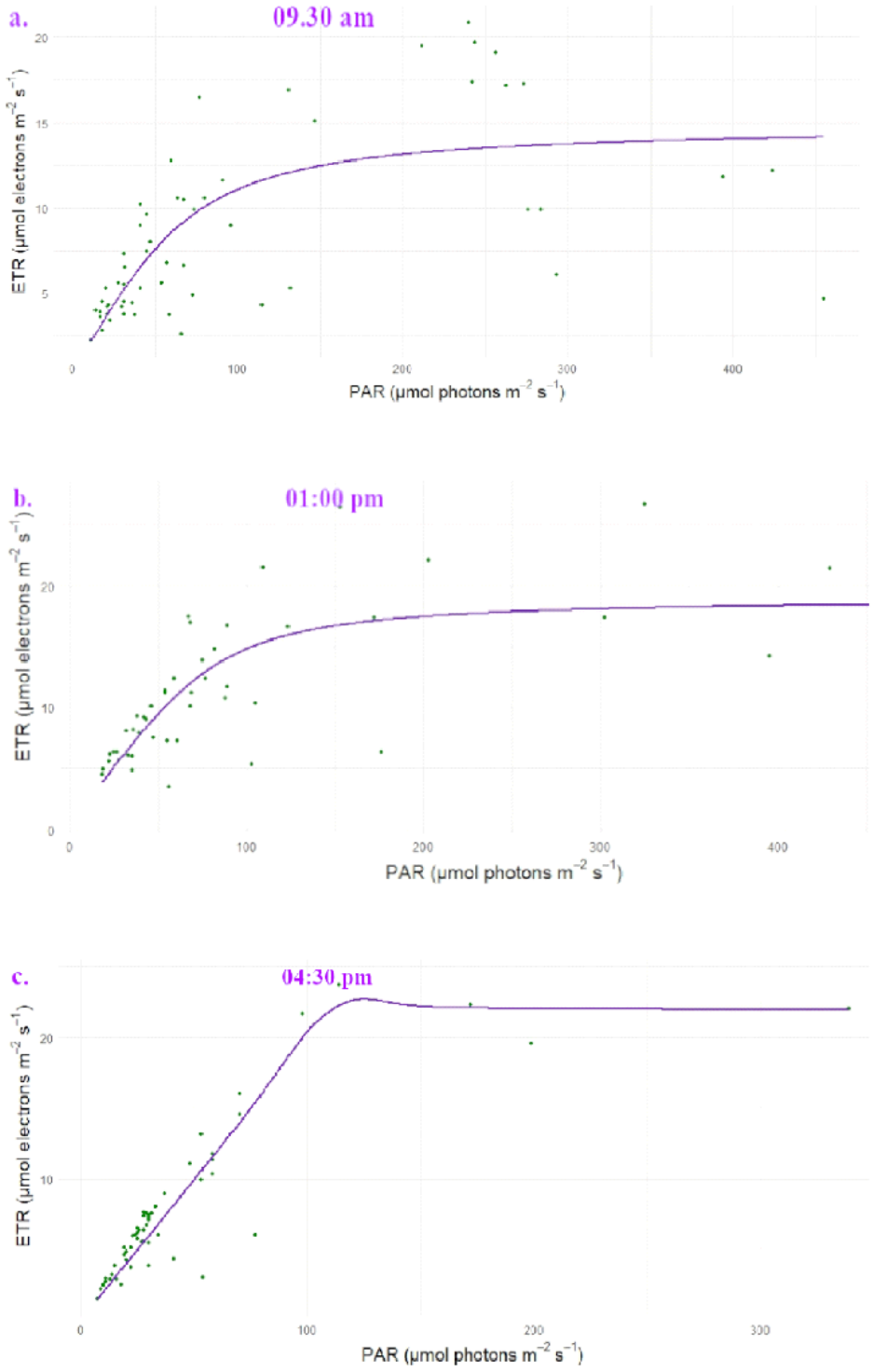

Figure 7

Light response curves for 2023 at different times of day; a) 09:30 am, b) 01:00 pm, c) 04:30 pm

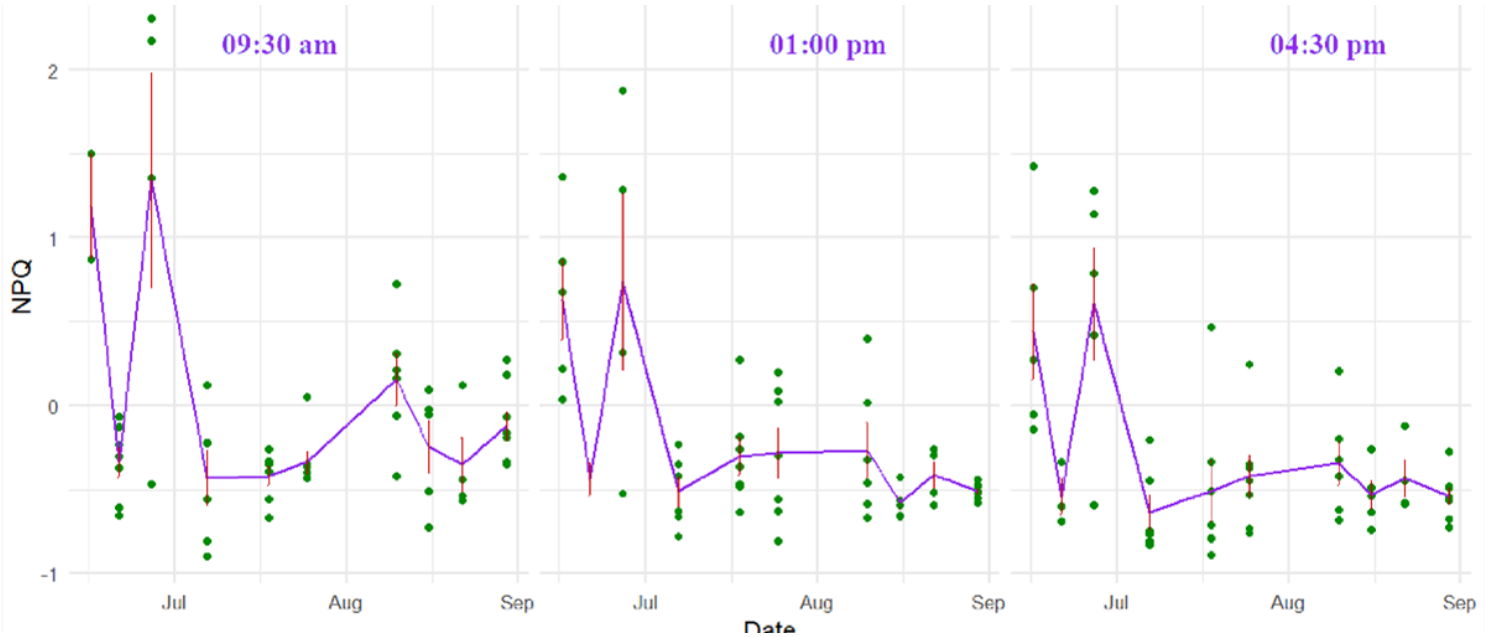

Figure 8

Standardised time series plot of NPQ during the 2023 season at different times of day.