### **Combined Effect of Light and Heat on Leaf Function**

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# ABSTRACT

As global temperatures rise due to climate change, understanding how plants respond to increasing heat and light stress is crucial for predicting ecosystem resilience. This study investigates the combined effects of light and heat stress on the thermal tolerance of five native California plant species, focusing on Photosystem II (PSII) efficiency, measured by the  $F_v/F_m$  ratio, under varying temperature and light intensities. The results reveal that high light intensity significantly reduces thermal tolerance in four of the five species. Notably, *Prunus fremontii* exhibited unique resilience by maintaining stable thermal tolerance across light conditions. These findings provide valuable insights into how climate extremes affect plant function and biodiversity, offering guidance for conservation strategies aimed at preserving native species in the face of rising environmental stressors.

Keywords: Light stress, Heat stress, Photosynthesis, PSII, Relative Water Content

#### INTRODUCTION

With global temperatures continuing to rise, plants face multiple stressors, including heat and light, which can impair critical functions such as photosynthesis. This study examines how simultaneous heat and light stress affect the photosynthetic efficiency and thermal tolerance of five native California plant species. Specifically, it focuses on Photosystem II (PSII) efficiency and the  $T_{50}$  metric—the temperature at which PSII efficiency is reduced by half. As California's average temperatures have risen significantly since the 19th century (OEHHA, 2018), understanding how native plants respond to these environmental changes is crucial for predicting their resilience and supporting biodiversity conservation (NASA, 2023). While individual effects of heat and light stress are well-researched, there is limited data on how they interact. Indeed, though often high temperatures are matched with high light levels in nature, most studies have investigated heat sensitivity responses of leaves under dark conditions (Curtis *et al.*, 2014, Sastry & Barua, 2017, Slot *et al.*, 2019, Slot *et al.*, 2021, Kunert *et al.*, 2022, Gauthey *et al.*, 2023, Münchinger *et al.*, 2023, Valliere *et al.*, 2024). This study aims to fill this gap by examining these stressors' combined effects on PSII efficiency, providing data essential for developing effective conservation strategies.

# METHODS

The research was conducted at the California Botanic Garden, supported by the USDA REEU grant. Five native California species were studied: *Salvia apiana* (white sage), *Cercocarpus betuloides* (mountain mahogany), *Ceanothus leucodermis* (chaparral whitethorn), Comarostaphylis diversifolia (summer holly), and *Prunus fremontii* (desert apricot). Branches were collected in the garden and transported in dark moist plastic bags to the lab and rehydrated overnight. The next morning, fully hydrated leaves were cut from the branches, placed in plastic bags, and exposed to water baths of varying temperatures and light conditions. Ten temperature treatments were chosen to range from 30°C to 58°C, and light conditions were set to low (<10 µmol m^(-2) s^(-1)) and high (>1500 µmol m^(-2) s^(-1)) intensities (Kalaji et al., 2014).

PSII efficiency was assessed using a fluorometer and calculating the  $F_v/F_m$  ratio, which measures photosynthetic health. After the exposure to temperature, leaves were left to recover in a

moist plastic bag under lab temperatures (20C) for 24h. We measured the  $F_v/F_m$  ratios of the recovered leaf using a fluorometer, to determine the degree of irreparable damage produced by the heat/ light. We constructed leaf thermal sensitivity curves for each species under the two light levels by plotting  $F_v/F_m$  vs. temperature. We fit a logistic function through each curve and extracted the temperature at which 50% of  $F_v/F_m$  had declined ( $T_{50}$ ). The  $T_{50}$  obtained under low vs high light for each species was compared by determining whether they overlapped in their 95% confidence intervals. To test whether leaves might be deprived of oxygen in a moist bag for 24h (and thus potentially impeded the recovery process), we tested recovery via two other methods, still ensuring leaves remained moist throughout the recovery process: 1) placing the leaf in a bag filled with moist paper towel, but open to the atmosphere, and 2) placing the leaf petiole in water with wet paper towels around the leaf (but not touching the leaf) and open to the atmosphere. We tested for a difference in recovery in  $F_v/F_m$  across recovery treatments using a two-way ANOVA.

#### RESULTS

The study found that high light intensity significantly lowered the  $T_{50}$  for four of the five species. Under high light conditions, thermal tolerance ( $T_{50}$ ) was reduced by 1 to 6°C compared to low light conditions, highlighting the impact of light on heat tolerance. *Prunus fremontii* was the only species that maintained stable thermal tolerance under varying light intensities, demonstrating unique resilience. Statistical analysis showed significant differences in  $T_{50}$  values between light treatments, with non-overlapping 95% confidence intervals. No significant differences were found across recovery treatments across species, and our data confirmed that recovery methods did not lead to dehydration, as relative water content (RWC) remained above 95% across all treatments.

#### DISCUSSION

These results emphasize the critical role of light intensity in exacerbating heat stress. The significant reduction in thermal tolerance under high light conditions for most species suggests that previous studies conducted under low-light or dark conditions may overestimate plant resilience. The stable performance of *Prunus fremontii* indicates that some species possess adaptive traits that allow them to withstand combined heat and light stress better than others. Future work should investigate the anatomical and biochemical drivers behind this light effect, and thermal resilience.

Previous studies suggest That reactive oxygen species (ROS) generated under high light and/or high temperature conditions may explain the differences in thermal tolerance. Indeed, increased ROS levels under high light can overwhelm antioxidative defenses, leading to greater damage of proteins in PSII and/or affect membrane fluidity for repair under heat stress (Yamamoto and Shimizu, 2016). The variability in responses among species points to the need for speciesspecific conservation strategies.

# CONCLUSIONS

This study provides valuable insights into the effects of combined light and heat stress on the thermal tolerance of native California plant species. Light intensity significantly lowers thermal tolerance in most species, but *Prunus fremontii*, a species native to the Mojave Desert, stands out for its lack of enhancement in thermal sensitivity when exposed to high light. These findings underscore the importance of considering light conditions when evaluating plant stress responses, as studies that do not account for light may overestimate thermal tolerance. Further research should investigate the biochemical and anatomical traits that enable some species to withstand combined stressors and explore recovery dynamics under natural environmental conditions. Such knowledge is essential for developing targeted conservation strategies to protect vulnerable species as climate change accelerates. Further, understanding the drivers behind thermal tolerance under different light environments can be valuable to help engineer more resistant crops under climate change.

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