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Physiological Effects of Disrupted Circadian Cycles in *Geranium oreganum*

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Introduction

Circadian rhythms govern an organism's internal clock and regulate processes such as gene expression, stomatal opening, and reproductive cycles in plants (1). Most organisms base circadian cycles upon patterns of sunlight and temperature changes and function most optimally upon an equinoctial day-night cycle with roughly equal periods of light and dark within a 24-hour period (2). Plants are especially sensitive to patterns of sunlight and understanding the response of plants to abnormal circadian rhythms can aid in agricultural development of areas where sunlight is at a premium.

Methods and Materials

Ten *Geranium oreganum* plants were selected from a pre-established greenhouse population. Each specimen had mature leaves measuring larger than 3 inches removed and were fertilized with Miracle Gro and Osmocote Plus. Specimens were arranged in a blocked greenhouse design and sorted randomly into treatments. Control plants were given an equinoctial day-night cycle with 12 hours of daylight and 12 hours of darkness each day while treatment plants experienced a blocked day-night cycle with alternating periods of 24-hour sunlight and 24-hour darkness. Daylight treatments were controlled via artificial growth lights and darkness treatments for the experimental group were administered in a sealed basement cupboard. Photosynthetic rates for light fleck tests and maximum conductance rate were measured via LI-COR 6400 (Lincoln, NE). Specific leaf area and minimum conductance rates were calculated using imageJ (NIH Bethesda, MD).

Results

Physiologically, the *Geranium oreganum* plants under treatment showed several differences to the control treatment plants. Both minimum and maximum conductance of water were affected with the control plants having a lower minimum conductance but a higher maximum conductance (figure 1). Treatment plants also had a much higher stomatal density (figure 3) than control plants but a lower specific leaf area (figure 2). A light fleck test was conducted on the plants which revealed that, when adapted to light, treatment plants and control plants behaved similarly to the initial 30 second burst of light. However, the second, longer burst of light during the test showed the treatment plants respond with a lower photosynthetic rate than the control plants as well as a lower photosynthetic rate than after the first fleck of light of the test (figure 4 and table 1).

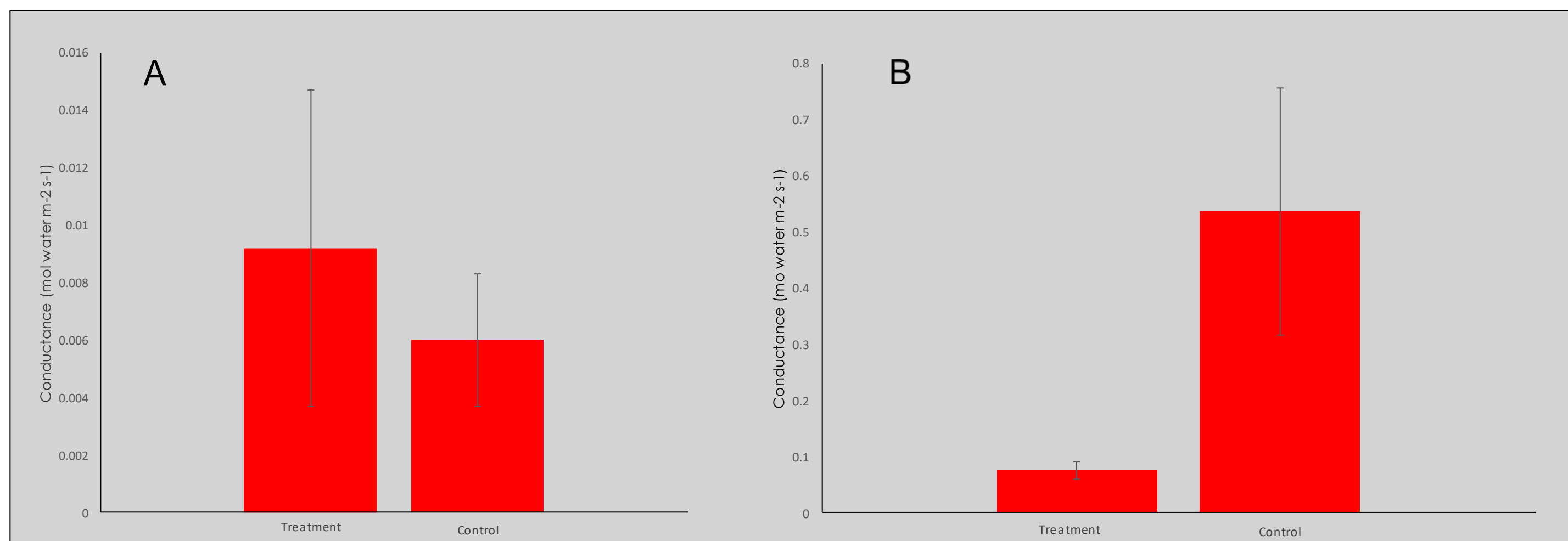


Figure 1. Minimum (A) and maximum (B) conductance in *Geranium oreganum*. Values are means +/- standard deviation. A) $t_9 = 1.19$, $p > 0.05$. B) $t_9 = -4.67$, $p < 0.05$.

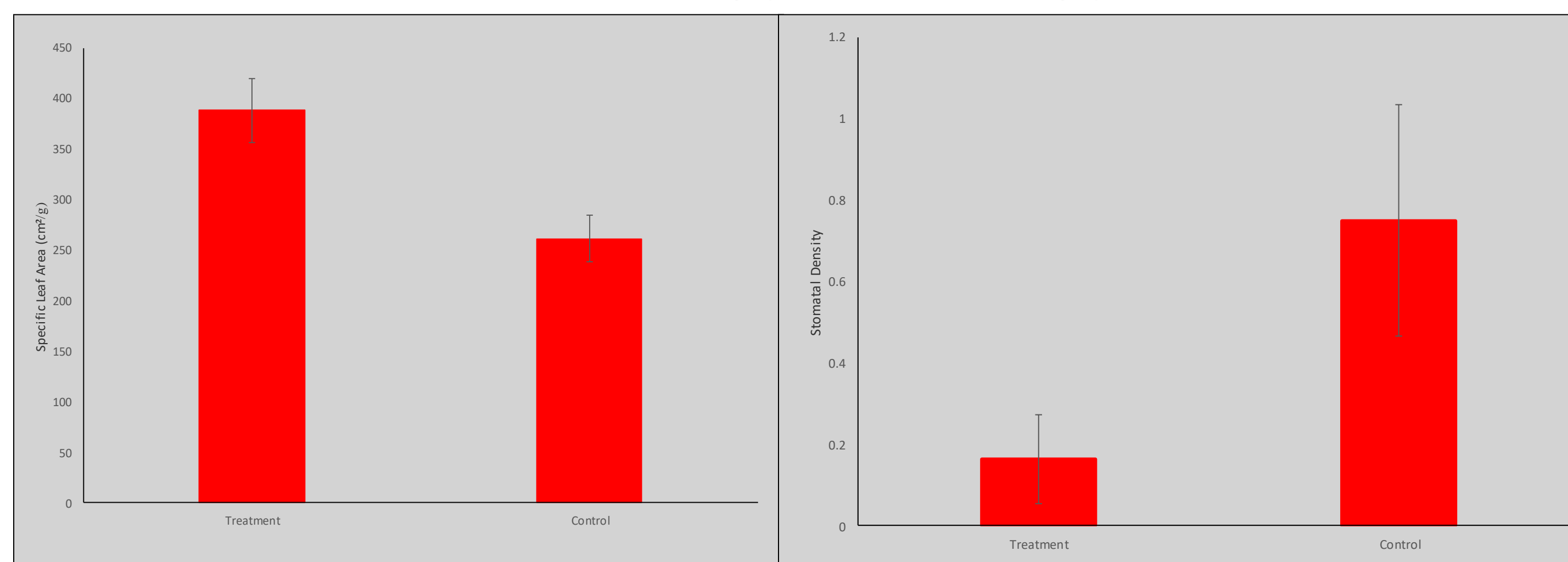


Figure 2. Specific leaf area. Values are means +/- standard deviation. $t_9 = 7.26$, $p < 0.001$.

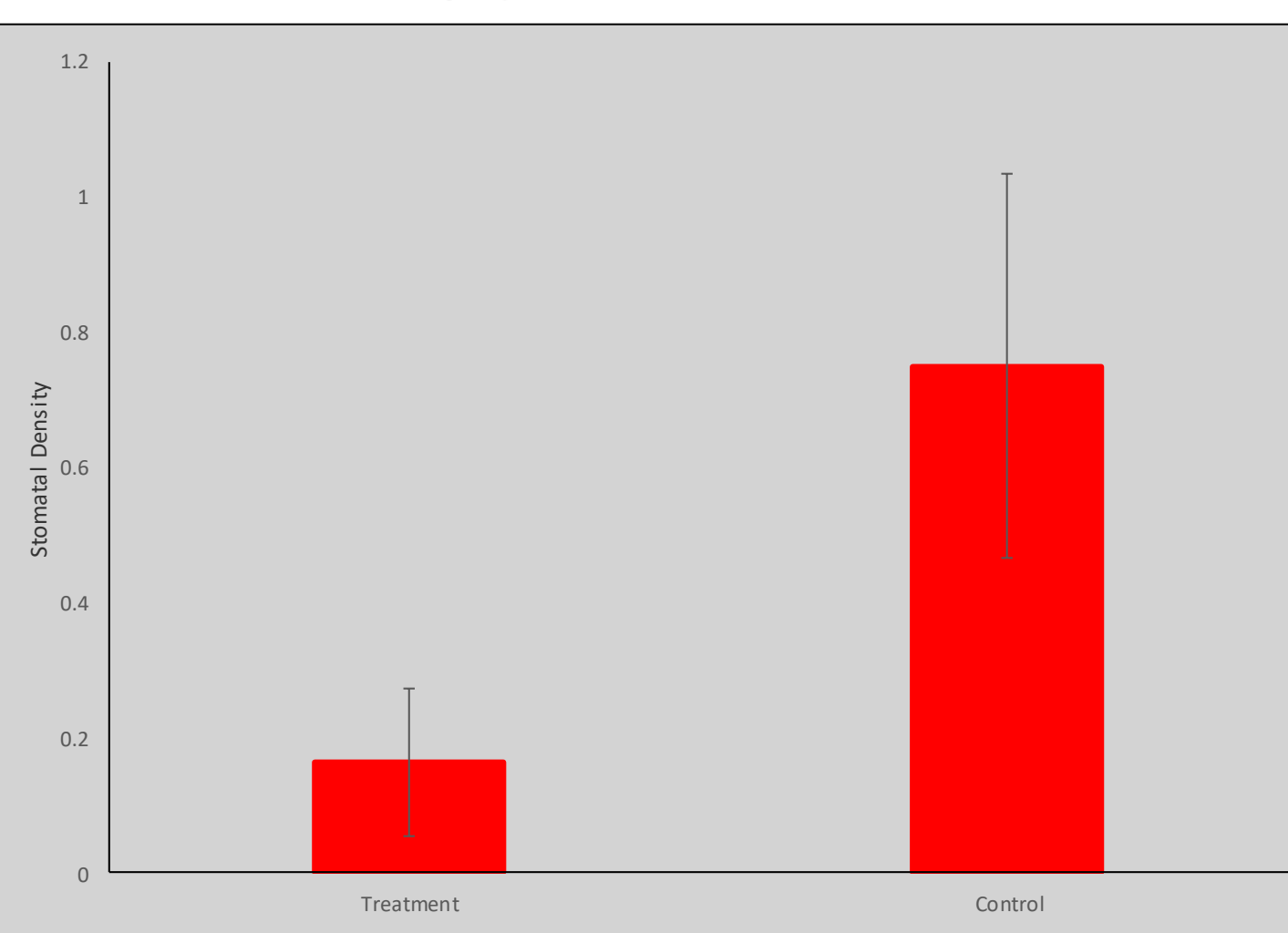


Figure 3. Density of stomata per square centimeter. Values are means +/- standard deviation. $t_9 = 4.30$, $p < 0.05$.

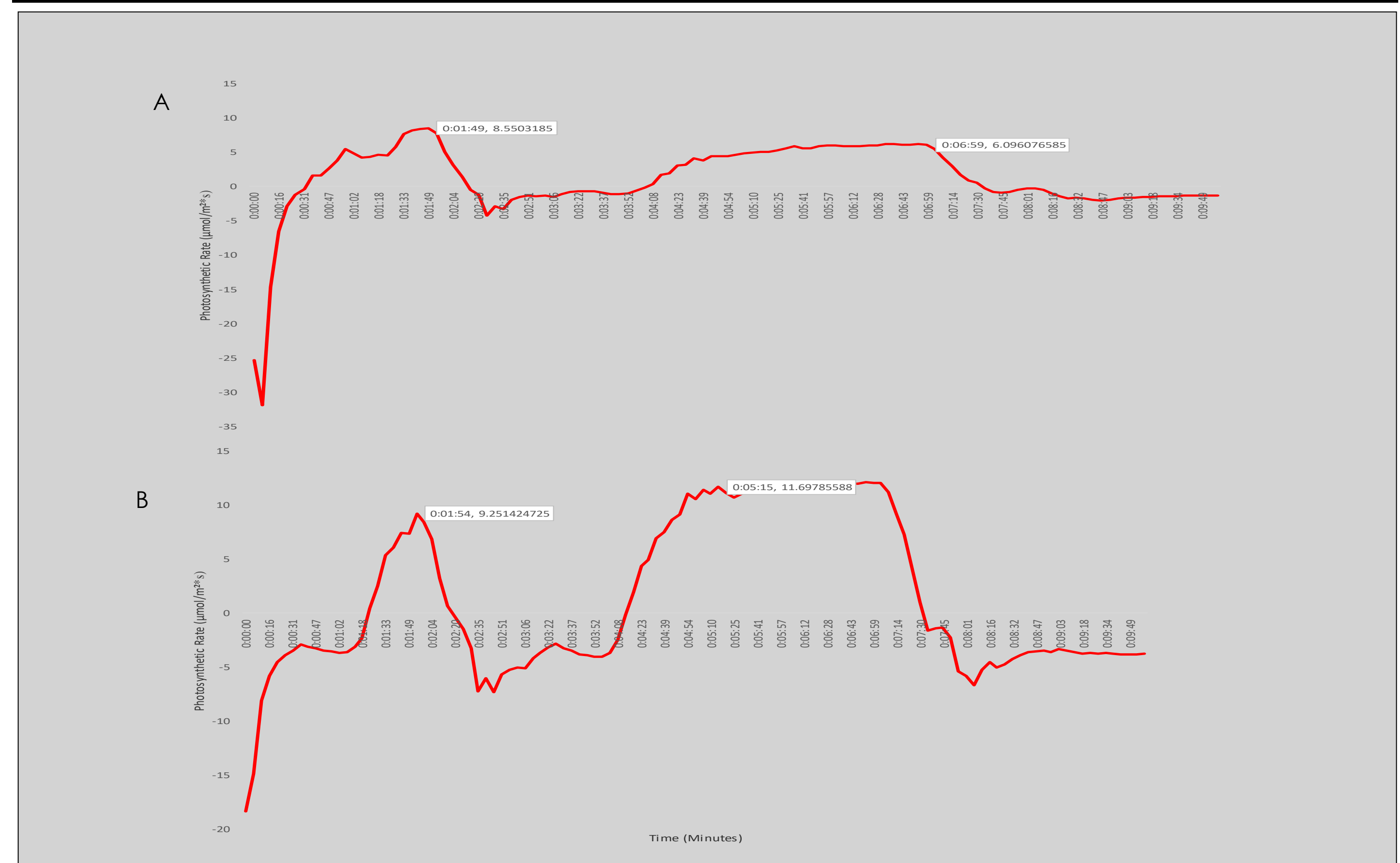


Figure 4. Average time to reach maximum photosynthetic rate after quick flecks of light in treatment plants (A) and control plants (B). Values are means +/- standard deviation.

	Dark Acclimated		Light Acclimated	
	Time to Peak (seconds)	max A	Time to Peak (seconds)	max A
Treatment	109	8.55	419	6.1
Control	114	9.25	315	11.7
	$t_4 = -1.27$	$t_4 = -0.22$	$t_4 = -0.11$	$t_4 = -5.81$
	$p > 0.05$	$p > 0.05$	$p > 0.05$	$p < 0.005$

Table 1. Maximum photosynthetic rate (max A) and time to reach max A after exposure to light flecks. Values are means.

Conclusions

Conductance (g) is known to fluctuate in plants during flowering and disruption of circadian rhythms can alter the reproductive cycle in plants (3 and 4). Lowered g is an indicator of flowering in some species and the control plants displayed a lower minimum g which could imply these specimens may be more fit for reproduction (4). Discrepancies in g between control and treatment plants could be an indicator that the treatment plants had stunted reproduction in response to the longer night period which could account for the narrow range between minimum and maximum g. These conclusions were not tested, however, and further study must be conducted to support this theory. Heightened maximum g in control plants could also be due to the heightened number of stomata control plants had compared to treatment plants. Treatment plants showed a higher specific leaf area (SLA) indicating these plants produced thin, broad leaves to capture the limited light available to them. The control plants' response to light flecks resulted in a classic curve expected from this type of experiment, however, the treatment plants responded to a second, prolonged period of light with a lowered photosynthetic rate than after the first burst of light. This could indicate that the plants are acclimating quickly to the dark period between flecks by shutting down photosynthesis but not acclimating as quickly to light. This could be for many reasons but is most likely an energy conservation technique for the plants since they are used to receiving 24 hours of light rather than needing to take advantage of quick bursts of light.

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