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EFFECTS OF BLUE AND GREEN LIGHT ON PLANT GROWTH
AND DEVELOPMENT AT LOW AND HIGH
PHOTOSYNTHETIC PHOTON FLUX

by

Michael Chase Snowden

A thesis submitted in partial fulfillment
of the requirements for the degree

of

MASTER OF SCIENCE

in

Plant Science

(Crop Physiology)

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2015

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ABSTRACT

Effects of Blue and Green Light on Plant Growth and Development
at Low and High Photosynthetic Photon Flux

by

Michael Chase Snowden, Master of Science

Utah State University, 2015

Major Professor: Dr. Bruce Bugbee
Department: Plant, Soils and Climate

The optimal combination of wavelengths of light (spectral quality) for single leaf photosynthesis has been well characterized, but spectral quality is not well characterized in whole plants in long-term studies. Here we report the effects of eight light spectra at two photosynthetic photon fluxes (200 and 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$) on dry mass, leaf area index and net assimilation of seven species in replicate 21-day studies. The combination of treatments allowed us to separately assess the effects of blue and green light fraction among species and PPF. At a PPF of 500, increasing blue light from 11 to 28 % significantly decreased dry mass in tomato, cucumber, and pepper, but there was no significant effect on soybean, lettuce and wheat. At a PPF of 200, dry mass significantly decreased only in tomato across the blue light range. Effects on leaf area paralleled effects on dry mass in all species at both PPFs, indicating that the effects of blue light on dry mass were mediated by changes in leaf area. Contrary to predictions of net

assimilation based on blue light response of single leaves, there was no evidence of decreasing net assimilation with increasing blue light. In contrast to the significant effect of blue light dry mass and leaf area, increasing green light fraction from zero to 30 % resulted in few significant differences. Contrary to several reports on significant green light effects on growth (both increases and decreases), we found no consistent effect of green light among species on growth, leaf area or net assimilation. Collectively, these results indicate significant differences among species in sensitivity to blue light and less sensitivity to green light, and that the effect of blue light on dry mass is primarily determined by changes in leaf area.

(71 pages)

PUBLIC ABSTRACT

Effects of Blue and Green Light on Plant Growth and Development at Low and High Photosynthetic Photon Flux

Michael Chase Snowden

Research in photobiology dates back over 200 years with studies using primitive light sources. This early research identified photoreceptors and action spectra for specific regions of the light spectrum that are paramount for photosynthesis as well as growth and development that are still topics of interest today.

Photobiological research has become an area of increasing interest since the introduction of light-emitting diodes which allow for evaluating endless combinations of light spectra. Red light-emitting diodes were the first to be introduced that had an electrical efficiency comparable to existing light sources. The research found that red light alone was not sufficient to promote normal plant growth and development in most species and that some blue light supplementation was needed. The amount of blue light required has been extensively studied with varying results.

The introduction of light emitting diodes has also allowed for studies of the effects of green light on plant growth and development. The influence of green light, similar to blue light, has resulted in varying conclusions, mainly regarding the importance of green light for photosynthesis.

This research will provide important information on optimal light quality for commercial greenhouse and controlled environmental food production.

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I would like to thank Dr. Bruce Bugbee for the opportunity to learn and conduct research in photobiology under his guidance and excellent mentorship. I would like to thank my committee members Dr. Dan Drost and Dr. Lance Seefeldt for their willingness to serve on my committee and provide valuable input to my research and thesis to make this degree the upmost quality it could be. I would also like to thank my cohort and amazing office mate Lance Stott for is wonderful friendship and laughter provided throughout this process, also Sandra Rhodes and Boston Swan for helping out with data collection and becoming good friends in the process, none of you will ever be forgotten. Special thanks to Alec Hay for his technical expertise in everything, well disguised pranks and good conversations. I am also extremely thankful for the friendship group that I have established during my time in Logan and their willingness and dedication to be there for me during this challenging time of graduate school and personal obstacles.

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LIST OF ACRONYMS

PPF, Photosynthetic Photon Flux

LEDs, Light Emitting Diodes

BL, Blue Light

GL, Green Light

RL, Red Light

RB, Red and Blue

RGB, Red, Green and Blue

DM, Dry Mass

LAI, Leaf Area Index

INTRODUCTION

Utilization of light by plants

Radiation is the source of energy for photosynthesis, and provides information for photomorphogenesis (Smith, 2013). Photosynthesis is driven by photosynthetically active radiation (400 to 700 nm). Pigments used by plants to capture photosynthetically active radiation for photosynthesis include chlorophyll a, chlorophyll b and a variety of accessory pigments, including carotenoids, which transfer absorbed energy to photosynthetic reaction centers (Nishio, 2000). Photosynthetic photon flux ($\mu\text{mol photons m}^{-2} \text{ s}^{-1}$) is used to measure photosynthetically active radiation and gives equal weight to all photons within the photosynthetically active radiation range (McCree, 1972b). However, not all wavelengths of photons are equally efficient in driving photosynthesis. Each pigment has a characteristic action spectrum, which is defined as the rate of physiological activity at each wavelength of light. The action spectra of chlorophyll a and b strongly absorb red light (RL) and blue light (BL), but weakly absorb green light (GL) (Nishio, 2000).

Experiments by Hoover (1937) on photosynthetic efficiency curves over the light spectrum served as the foundation for the relative quantum efficiency curves established by McCree (1972a), which is cited in most current research related to photobiology and light quality. The relative quantum efficiency curve indicates that RL (600 to 700 nm) is 25 to 35 % more efficient than BL (400 to 500 nm) and 5 to 30 % more efficient than GL (500 to 600 nm) in driving photosynthesis (Inada, 1976; McCree, 1972a). However, this curve was measured with single leaves, at a low photosynthetic photon flux (PPF) and

over short intervals. Therefore, it may not be representative of whole plants or plant communities grown at high PPF under mixed colors of light. It also indicates only the photosynthetic efficiency and not the combined effects of photosynthesis and development.

Photomorphogenesis is defined as light-mediated development, and is regulated by the light perception network, which is composed of at least three classes of photoreceptors: phytochromes, cryptochromes, and phototropin (Briggs and Olney, 2001). Phytochromes are sensitive to red and far-red radiation, cryptochromes to blue and ultraviolet-A, and phototropins to blue, green and ultraviolet-A (Casal, 2000). In *Arabidopsis thaliana*, five phytochromes (Phy A through Phy E), two cryptochromes (Cry 1 and Cry 2), and two phototropins (Phot 1 and Phot 2) have been identified (Quail, 2002). The activity of each photoreceptor varies with developmental stage (Sullivan and Deng, 2003). Interactions among photoreceptors can be synergistic or antagonistic, depending on the light signals received by the plant (Casal, 2000). Most if not all of the photoreceptors in *A. thaliana* are likely conserved in other plants species (Chaves et al., 2011). The activity of these photoreceptors can be manipulated by altering the light spectra (Folta and Childers, 2008; Stutte, 2009).

History of photobiological research

Photobiological research dates back more than 200 years. Wassink and Stolwijk (1956) provide an excellent review of early photobiological research involving light quality effects on plant growth and development. The authors discuss research using “Senebier domes”, which are double walled glass covers filled with colored fluids put

over plants to provide spectral filters for research (von Sachs, 1864). Interestingly, that research, conducted with electric lighting sources was able to manipulate the light spectra to test plant growth and development with narrow wavelengths of light quality, which is what research with LED technology is trying to do today. The review by Wassink and Stolwijk (1956) indicates that it was during this time that the photoreversibility of red and infrared light was discovered, which led to the presence of the red light photoreceptor, phytochrome. The review also discussed a study by Crocker (1948) demonstrating the importance of the red and blue regions of the spectrum in which the red region results in spindly, elongated and curled leaves, while the blue region promoted compact growth and flat expansion of leaves.

The development of LEDs has made it possible to manipulate spectral quality in ways that have been difficult with conventional electric light sources (Morrow, 2008). As such, LEDs have been used to confirm the role and importance of light quality (Massa et al., 2008) and the ability to strategically manipulate plant growth and development (Folta and Childers, 2008; Stutte, 2009). Interestingly, although RL is the most efficient in driving photosynthesis, alone it does not promote normal development (Barta et al., 1992; Bula et al., 1991; Goins et al., 1997; Hoenecke et al., 1992; Kim et al., 2005). Supplementation with BL is necessary to mitigate the shade avoidance responses induced by RL and produces compact plant shape with shorter stems and decreased leaf area resulting in decreased growth (dry mass) (Dougher and Bugbee, 2001; Kim et al., 2005; Yorio et al., 2001).

Effects of blue light

The amount of BL required to promote normal development varies among species; furthermore, developmental parameters within a species potentially respond to either the absolute ($\mu\text{mol m}^{-2} \text{s}^{-1}$ of photons between 400 and 500 nm) or the relative (percent of total PPF) amount of BL (Cope and Bugbee, 2013; Dougher and Bugbee, 2001; Hoenecke et al., 1992). Wheeler et al. (1991) were the first to propose that plant developmental response to blue light was dependent on absolute BL rather than the relative amount of BL for stem length in soybean. The authors concluded that approximately $30 \mu\text{mol m}^{-2} \text{s}^{-1}$ of BL was required to inhibit stem and internode elongation. Yorio et al. (1997) produced similar findings for wheat, potato, soybean, lettuce and radish which were shown to require a minimum of $30 \mu\text{mol m}^{-2} \text{s}^{-1}$ of BL for normal growth and development. In contrast, Cope and Bugbee (2013) indicated that $50 \mu\text{mol m}^{-2} \text{s}^{-1}$ or 15 % BL (whichever is greater depending on the PPF) is likely required to promote normal development in most species. Cope and Bugbee (2013) also concluded that some parameters for soybean, radish and wheat are better predicted by relative BL. These studies identify approximate absolute BL requirements that range between 10 to 15 % BL from the total PPF produced by the light sources tested. This indicates the need for further studies to determine if absolute or relative BL is the better predictor of plant growth and development.

Studies on light quality using LEDs to assess plant growth and development began with RL supplemented with BL of various sources (Bula et al., 1991; Goins et al., 1997; Hoenecke et al., 1992) and has progressed into evaluating ratios of RL and BL

(Hernández and Kubota, 2015; Son and Oh, 2013) or percentage of BL from varying light quality treatments (Cope et al., 2014). Cope et al. (2014) indicated that growth (dry mass) and leaf area increased when BL was added to a pure RL source, and increased up to 15 % BL for lettuce, radish and pepper. Similarly, Hernández and Kubota (2015) found an initial increase in leaf area and dry mass for cucumber from 0 to 10 % BL, however both parameters began decreasing with more than 10 % BL. These conclusions are supported by earlier studies by Bula et al. (1991) and Hoenecke et al. (1992) on lettuce using red LEDs supplemented with 10 % fluorescent BL. Yorio et al. (2001) also found decreased dry mass production under pure RL for spinach, radish and lettuce and that while dry mass was increased with the addition on 10 % BL added by fluorescent lamps the results were still lower than a fluorescent white control. Goins et al. (1997) studied wheat under identical light treatments to Yorio et al. (2001) and found that wheat seed yield was higher in the RL with 10 % BL compared to RL alone and was comparable to a white light control. Dougher and Bugbee (2004) described histological effects of BL on plant growth and development for lettuce and soybean and found that BL decreased cell expansion in etiolated plants and inhibited cell division in the stems of soybean. Leaf area was also reduced with increasing BL above 6 % for soybean due to decreased cell expansion.

In contrast to the previously mentioned studies, Son and Oh (2013) showed that biomass and leaf area were higher for both red and green lettuce in the zero BL treatment. These differences could be attributed to cultivar and cultural practices, but it is unclear why their findings do not follow the same trend as the other studies. Johkan et al. (2010)

studied lettuce seedling quality and found that adding BL to RL and pure BL resulted in compact morphology and promoted growth after transplanting compared to RL alone. Chen et al. (2014) conducted a similar study in which RL and BL LEDs were individually added to a fluorescent light source and compared to pure RL, pure BL and a combination of RL and BL. Chen et al. (2014) concluded that RL in combination with fluorescent light promoted seedling growth. The combination of BL added to fluorescent light improved growth during the maturation period compared to pure BL, RL and fluorescent light treatments for lettuce. Collectively, these studies indicate that the response to BL is species specific, and that sole source RL decreases growth and that 5-15 % BL is sufficient to promote optimal growth.

Numerous studies have examined the effects of increasing BL on photosynthetic efficiency. Goins et al. (1997) was one of the first studies to identify that BL increased net leaf photosynthesis compared to RL alone in wheat. Hogewoning et al. (2010a) found that only 7 % BL doubled the photosynthetic capacity over RL alone, and that photosynthetic capacity continued to increase with increasing BL up to 50 % BL for cucumber. Terfa et al. (2013) found that LEDs with 20 % BL compared to high pressure sodium lamps with 5 % BL increased leaf mass per unit leaf area and increased photosynthetic capacity by 20 %. In contrast, studies by Ouzounis et al. (2014) and Ouzounis et al. (2015) showed decreases or no effect on photosynthesis with increasing BL for roses, chrysanthemums and campanulas and lettuce. Ouzounis et al. (2015) found a decrease of quantum yield by photosystem II and increase of non-photochemical quenching with increasing BL in red lettuce. Wang et al. (2014) also net photosynthetic

rate increased with BL for cucumber and that BL was essential for optimized photosynthetic performance.

Most studies have used chlorophyll fluorescence to determine photosynthetic efficiency using the method described in Genty et al. (1989), however a classic method of determining photosynthetic efficiency uses crop growth rate ($\text{g m}^{-2} \text{d}^{-1}$) as described in Leopold and Kriedemann (1975) and Hunt (1982) as:

$$\text{CGR} = \text{NAR} \times \text{LAI}$$

where NAR is the net assimilation rate (grams of dry mass per m^2 of leaf) and LAI is the leaf area index (m^2 of leaf per m^2 of ground). The ratio of growth to leaf area index provides a measure of net assimilation integrated over time. Because photon flux was constant at either 200 or 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ this is a measure of photosynthetic efficiency.

This method was used by Poorter and Remkes (1990) for analysis of 24 wild species and concluded that it was not net assimilation rate, but LAI that was a better predictor of growth. Bullock et al. (1988) studied corn growth in equidistant and conventional plant-spacing patterns and concluded that crop growth rate was higher in the equidistant treatments due to higher LAI rather than net assimilation rate. Klassen et al. (2003) also used this method and produced similar findings. Goins et al. (2001) used crop growth rate to study the effects of different wavelengths of RL and a constant BL level (8-9 % BL) from LEDs on lettuce and radish and concluded that increased incident radiation capture resulted in increased growth not higher individual leaf photosynthetic rates. Hogewoning et al. (2010b) also found similar findings in cucumber grown under artificial solar, fluorescent and high pressure sodium lamps. Growth of cucumber was at

least one and a half times greater under the artificial solar treatment which was related to more efficient light interception not photosynthesis.

Effects of green light

Similar to BL, GL may play a major role in controlling plant development in orchestration with RL and BL (Folta and Maruhnich, 2007), although its role is likely more important at low light conditions found within a canopy or high plant densities (Wang and Folta, 2013). Sun et al. (1998) found that both RL and especially BL drive CO₂ fixation primarily in the upper palisade mesophyll (adaxial portion of the leaf) while GL penetrates deeper and drives CO₂ fixation in the lower palisade and upper spongy mesophyll (abaxial portion of the leaf). Broadersen and Vogelmann (2010) studied leaf cross sections for chlorophyll fluorescence imaging to show that diffuse and direct RL, GL and BL penetrate leaves differently. For all three colors, direct light penetrated more deeply than diffuse light, and for both direct and diffuse light, GL penetrated much deeper than RL and BL. Accordingly, once both the upper part of individual leaves and the upper canopy as a whole are saturated by RL and BL, additional GL is likely more beneficial for increasing whole plant photosynthesis than RL or BL (Nishio, 2000). This effect was experimentally confirmed by Terashima et al. (2009) who reported that at high PPF, GL drives leaf photosynthesis more efficiently than RL and BL. Thus, whole plant photosynthesis could be increased by GL in two ways: one, by increasing total photosynthesis in the individual leaves, and two, by transmitting to lower leaf layers.

Whole plant studies suggest that supplemental GL may improve plant growth. Kim et al. (2004a) reported that too much GL (51 %) or too little (0 %) decreased growth,

while roughly 24 % was ideal. Johkan et al. (2012) studied lettuce growth at three PPFs (100, 200, and 300 $\mu\text{mol m}^{-2} \text{s}^{-1}$) using three wavelengths of green LEDs (peak wavelengths of 510, 520, and 530 nm) and cool white fluorescent controls at all three PPFs. Interestingly, the lettuce plants responded differently to both the three PPFs and the three wavelengths of GL. As PPF decreased and the wavelength of GL increased, the lettuce plants exhibited an increased shade-avoidance response. It is difficult to say whether the cause for this response in Johkan et al. (2012) was due to the reduction of blue wavelengths as the green wavelength increased from 510 to 530 nm or due to the change in green wavelengths, or both. Johkan et al. (2012) also reported that the lettuce grown under the cool white fluorescent lamps at all three PPFs developed more normally than the lettuce grown under all three wavelengths of GL. Furthermore, growth was consistently higher in the cool white fluorescent compared to the GL treatments except for the 510 nm treatment at 300 $\mu\text{mol m}^{-2} \text{s}^{-1}$. These results are consistent with findings of Kim et al. (2004b) by showing that too much GL is detrimental; however, unlike Kim et al. (2004b), this study provides no lower limit for the optimal GL level since no treatments containing 0 % GL were used as a control. Nevertheless, Johkan et al. (2012) clearly showed that, although complex, there are distinct and even predictable interactions between wavelength and PPF. Because the highest PPF used was only 300 $\mu\text{mol m}^{-2} \text{s}^{-1}$, additional studies with pure GL need to be carried out at higher PPFs. Hernández and Kubota (2015) studied the effects of 28 % GL added to a RL and BL source on growth and development of cucumbers and concluded that the addition of GL had no effect. The 24 % GL level in Kim et al. (2004b) increased lettuce growth whereas

the 28 % GL in the Hernández and Kubota (2015) did not increase cucumber growth.

While there are differences in the experimental setups between the two studies, including light level, light type, and species type, this suggests that response to GL may be species specific.

Effects of light intensity

Light intensity is another important parameter affecting plant growth and development. Most research using LEDs have used light levels less than $200 \mu\text{mol m}^{-2} \text{s}^{-1}$. For some plant species this can induce shade avoidance responses, which are described by Smith and Whitelam (1997) as including stem and petiole elongation, decreased pigmentation, reduced leaf area, and a reduction in dry mass. Also some studies conducted on light quality did not have consistent intensities among treatments that introduce confounding factors and possibly inaccurate conclusions about light quality effects (Lefsrud et al., 2008)

Purpose for proposed research

Hernández and Kubota (2015) indicate the need for research at higher PPF and over more species in order to determine an optimal spectral quality using LEDs. Photobiological research using LEDs is also providing the opportunity to challenge previous understandings of photosynthetic efficiency defined by McCree (1972a) and Inada (1976) in which measurements on single leaves, for short durations and at low intensities can be extrapolated to whole canopies under sole-source-high-intensity light spectra. This indicates the need to refine theories that more accurately predict the effects of increasing BL and GL on plant growth.

In this study we sought to further elucidate an optimal spectral quality for plant growth and development. The objectives for this study were to compare the photosynthetic and morphological effects of broad spectrum and monochromatic light.

Determine if changes in blue light percentage affect plant growth and development.

Determine if changes in green light percentage affect plant growth and development.

Determine interactions between light quality and light intensity. Determine interactions between species and light quality.

MATERIALS AND METHODS

Light treatments

The experimental system included 16 chambers (eight arrays of LEDs at two PPFs [200 and 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$]). The LED arrays were: warm, neutral, and cool white (Multicomp; Newark, Gaffney, SC), monochromatic green, monochromatic blue, monochromatic red, and combinations of red and blue (RB) and red, green, and blue (RGB) (Luxeon Rebel Tri-Star LEDs; Quadica Developments Inc., Ontario, Canada). Chambers measured 19.5 x 23 x 30 cm (13455 cm^3) and the internal walls were lined with high-reflectance Mylar® (Fig. 1).

Measurements of photosynthetic photon flux (PPF), phytochrome photoequilibrium (Sager et al., 1988) and the fraction of blue (400 to 500 nm), green (500 to 600 nm) and red (600 to 700 nm) light in all LED treatments at the were made using a spectroradiometer (model PS-200; Apogee Instruments, Logan UT)(Table 1).

The spectral trace of each LED array is shown in Fig. 2. The PPF was maintained constant relative to the top of the plant canopy using a quantum sensor (LI-188B; LI-COR, Lincoln, NE) calibrated for each treatment against the spectroradiometer. The PPF of each chamber was manipulated by adjusting the electrical current sourced to each LED and the variability averaged was less than 1 % over the study. The photoperiod was 16-h day/8-h night for each study.

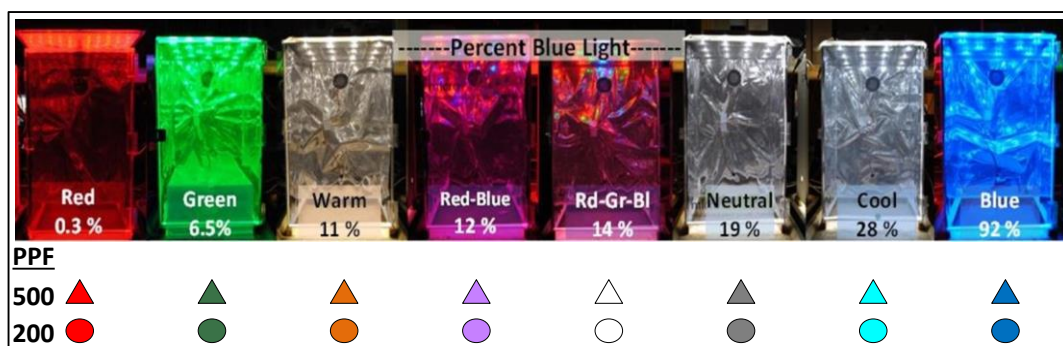


Fig. 1. The eight LED spectra and corresponding percent BL for each treatment. Symbols correspond to the color for each treatment and shape represents the two PPFs (200 and 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$). Symbol shape and color are consistent in all figures.

Table 1

Spectral characteristics of the eight treatments. Phytochrome photoequilibrium (PPE) as described by Sager et al. (1988). Blue, green and red values indicate percent of total PPF (400 to 700 nm).

| Light quality Parameter | SPECTRAL TREATMENT | | | | | | | |
|----------------------------|--------------------|-------|-------|------|-------|----------|-------|------|
| | Red | Green | Warm* | RB† | RGB*† | Neutral* | Cool* | Blue |
| % UV-A | 0.03 | 0.17 | 0.09 | 0.01 | 0.11 | 0.13 | 0.17 | 0.18 |
| % Blue | 0.27 | 6.52 | 10.8 | 12.0 | 13.7 | 19.4 | 27.5 | 92.0 |
| % Green | 1.63 | 92.5 | 41.0 | 1.70 | 22.9 | 45.6 | 48.0 | 7.77 |
| % Red | 98.1 | 0.96 | 48.2 | 86.3 | 63.4 | 35.0 | 24.5 | 0.22 |
| PPE | 0.89 | 0.83 | 0.84 | 0.89 | 0.89 | 0.84 | 0.83 | 0.58 |

*To minimize confounding spectral effects, only these four RGB, cool, warm and neutral white treatments were used in the blue light analysis.

†To minimize confounding spectral effects, only these three RGB, cool, warm and neutral white treatments were used in the green light analysis.

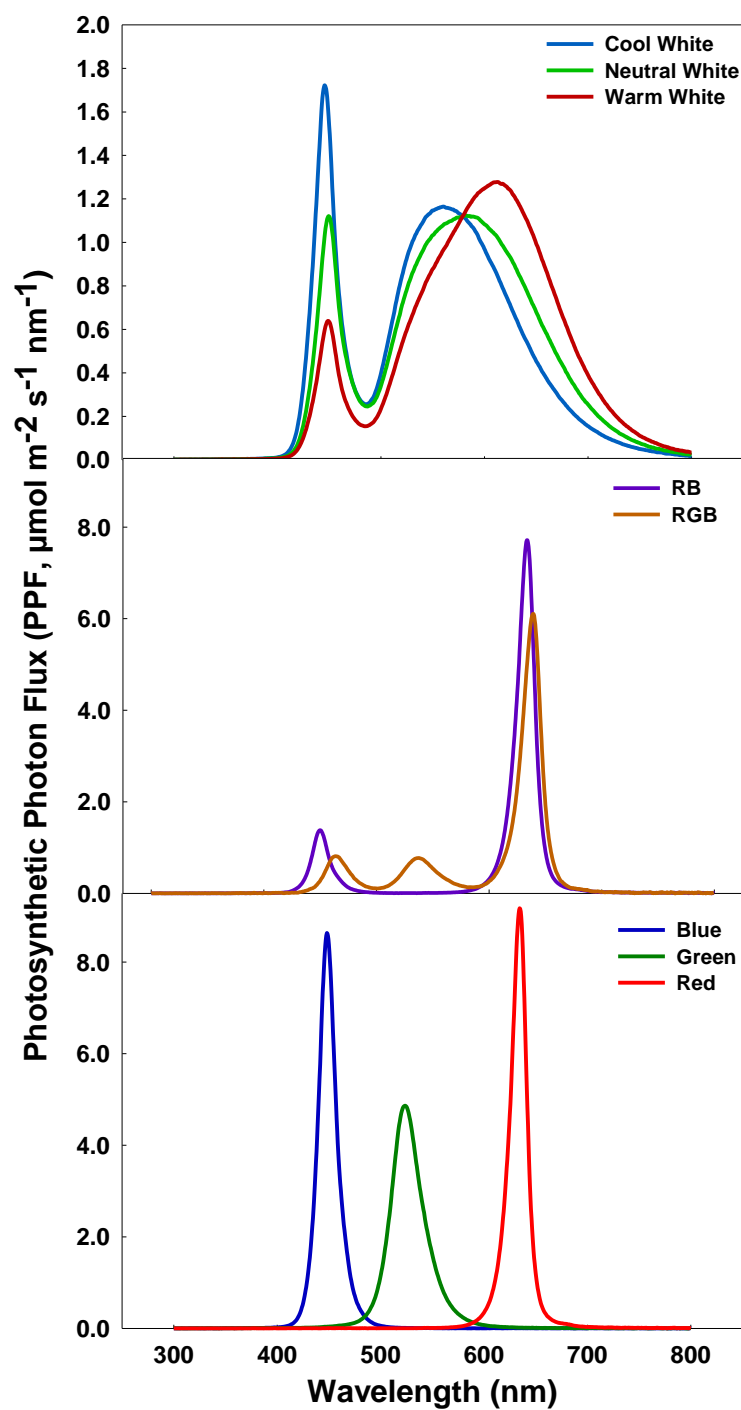


Fig. 2. Spectral distributions of all eight LED treatments, including: the three types of white LEDs, the red + blue (RB) and red + green + blue (RGB) LEDs, and the red, green, and blue monochromatic LEDs. Variation in the spectral distribution between the 200 and 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ treatments was negligible.

Environmental conditions for all growth chambers were controlled to maintain the same temperature, CO₂, and relative humidity. Type-E thermocouples connected to a data-logger (model CR1000, Campbell Scientific, Logan UT) were used to continuously monitor temperature at the top of the plant canopy. To avoid partial shading of the plants, the thermocouples were not shielded; if shielded, our measurements indicate that recorded temperatures would have been reduced by about 0.5°C in each treatment. Average temperature differences among chambers were less than 0.2°C. Relative humidity was measured using a relative humidity probe (model HMP110; Vaisala Inc, Finland) and CO₂ concentration was measured using a CO₂ probe (model GMP222; Vaisala Inc, Finland). Chambers were well ventilated to allow uniform conditions for CO₂ and relative humidity among the treatments. Relative humidity was approximately 30 % and CO₂ was approximately 430 μmol mol⁻¹.

Plant material and cultural conditions

Lettuce (*Lactuca sativa*, cv. 'Waldmann's Green'), cucumber (*Cucumis sativa*, cv. 'Sweet Slice'), wheat (*Triticum aestivum* L. cv. 'USU-Apogee'), tomato (*Lycopersicon lycopersicum* cv. 'Early girl'), soybean (*Glycine max*, cv. Hoyt) and radish (*Raphanus sativus*, cv. 'Cherry Belle') seeds were pre-germinated in a germination box until radicle emergence. Upon radical emergence, eight uniform seeds were transplanted to root modules measuring 15 x 18 x 9 cm (L x W x H; 2430 cm³), except wheat in which twelve seeds were transplanted. Root modules were filled with horticultural grade soilless media (1 peat: 1 vermiculite by volume) and 5 g of uniformly mixed slow-release fertilizer (16N-2.6P-11.2K; Polyon® 1 to 2 month release, 16-6-13). The media was watered to

excess with a complete, dilute fertilizer solution (100 ppm N; Scotts® Peat-Lite, 21-5-20; EC = 100 mS per m), and allowed to passively drain. This fertilizer solution was applied as needed to maintain ample root-zone moisture (every 2-3 days). The slow-release fertilizer and nutrient solution helped maintain leachate electrical conductivity measurements between 100 and 150 mS per m (1.0 to 1.5 mmhos per cm; 1 to 1.5 dS per m).

Pepper (*Capsicum annuum*, cv. 'California Wonder') seeds were pre-germinated in a germination box for 7 days, and two pre-germinated seeds with emerging radicles were transplanted into small pots measuring 8 x 8 x 7 cm (448 cm³). The modification of the cultural conditions for pepper from the other species was done to study the effects of light quality on growth stage. The pots were filled with soilless media identical to that used for the other species; however, due to the smaller volume of the pots, only 1 g of slow-release fertilizer was incorporated into each individual pot. The pots were then watered to excess with the same dilute fertilizer solution and allowed to passively drain. After planting, the pots were placed in a growth chamber (130 x 56 x 108 cm; 0.79m³) with an average PPF of 300 $\mu\text{mol m}^{-2} \text{s}^{-1}$ provided by cool white fluorescent lamps and day/night temperature set-points of 25/20 °C. Seedlings emerged within two days and the cotyledons were allowed to fully expand. Plants remained in the growth chamber for 33 days after emergence and the least uniform of the two germinated plants was removed. After 33 days after emergence, 48 of the most uniform plants were randomly assigned to 16 groups of four plants and were transplanted into root modules with the same dimensions as those used for the other species.

Plant measurements

All species were harvested 21 days after emergence, except cucumber and pepper which were grown for 14 and 54 days after emergence respectively. Termination at these time intervals was done in order to mitigate confounding factors between treatments caused by overlapping of plants in each chamber. Measurements were made on all four individual plants in each light treatment for each species except wheat. Wheat measurements were taken over all plants collectively per treatment. At harvest, relative leaf chlorophyll concentration (measured as chlorophyll content index) was measured with a chlorophyll meter (CCM-200; Opti-Sciences Inc., Hudson, NH). Measurements were taken on the third set of true leaves in lettuce, cucumber, wheat, tomato and radish, and the topmost, fully expanded leaves in pepper. Readings of chlorophyll content index were converted to absolute chlorophyll content using equations in Wellburn (1994). For all seven species at harvest, stem and leaf fresh mass, stem length, and number of leaves per plant was collected. Total leaf area was also measured using a portable leaf area meter (model LI-3000; LI-COR, Lincoln, NE). Once harvested, stems and leaves were separated and dried for 48 hours at 80°C for dry mass (DM) determination. Root mass was not measured. From the above measurements, specific leaf area (m^2 of leaf area g^{-1} of leaf DM), chlorophyll content ($\mu\text{mol m}^{-2}$ of leaf area) and net assimilation (g of DM per m^2 leaf area, which is an effective estimate of photosynthetic efficiency) were determined for all species.

Statistical analysis

There were three replicate studies for each species. Statistical analysis of the above growth parameters was conducted among species, at each light intensity, using a regression only the four treatments with RGB, cool, warm and neutral whites (Cool, Warm, Neutral, RGB) on a BL basis and the RB, RGB and warm treatments on a GL basis with the PROC-REG package in SAS (version 9.3; Cary, NC, USA). The significance level was set at $p = 0.05$.

RESULTS

An overhead view of one example replicate of cucumber for all treatments at both light levels is shown in Fig. 3. The treatments are arranged in order of increasing BL from left to right. Plants at both light levels grew well in the monochromatic green treatments, which is surprising due to the absorption of chlorophyll a and b predominately in the red and blue regions of the light spectrum. This indicates the dynamic ability of plants to adapt to the light environment. The chlorophyll concentration was reduced at the higher light level. It is difficult to visually distinguish differences in leaf area between treatments at the lower light level but leaf area was reduced in the red and blue treatments at higher light level. Visually, monochromatic blue light at the high light level overall decreased leaf area, compared to the multi-wavelength treatments (RB, RGB, cool, neutral and warm white). These visual results for cucumber suggest that there is a more pronounced response to the light treatments at the higher light level compared to the lower light level and the data for chlorophyll and leaf area support the visual observations. The monochromatic red and green treatments produced the lowest chlorophyll at both light levels, but increased with increasing BL up to 28 % BL (cool white treatment) and slightly decreased in the monochromatic blue treatment. Leaf area was lowest in the monochromatic red treatment and with the addition of 6 % BL in the monochromatic green treatment produced the highest leaf area out of all the treatments, additional BL significantly decreased leaf area. Visually this is more apparent at the higher light level than the lower light level.

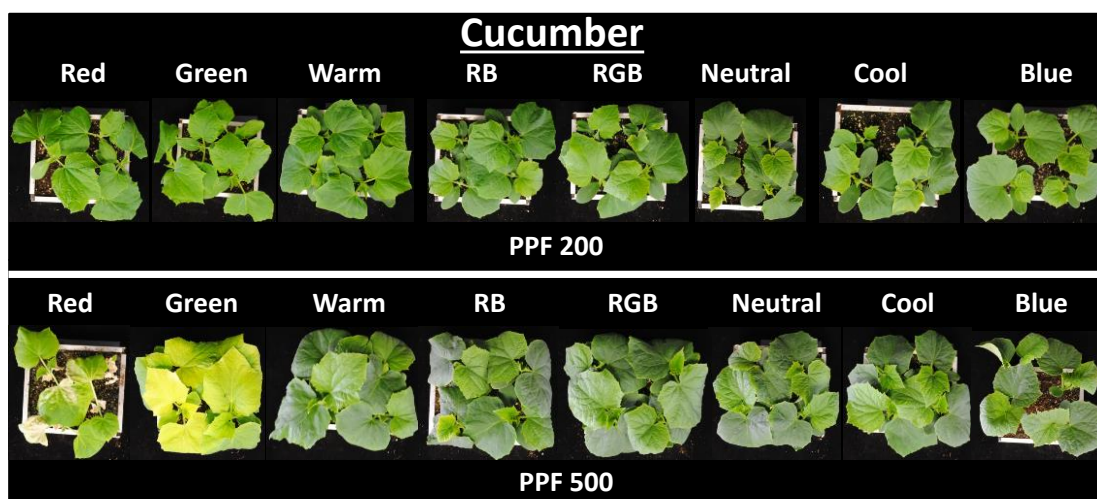


Fig. 3. Overhead view of cucumber at 21 days after emergence for all eight LED treatments at both light intensities. Note the stark contrast in coloration with the green and red treatments at the 500 PPF level, which contained the least amount of BL.

Statistical analysis

To mitigate confounding factors for the effect of BL, statistical analysis included only the four treatments (RGB, cool, warm and neutral white) (Fig. 1) that had comparable red and green light (Table 1). The RB treatment was not included due to the low GL with this treatment (0.71 %) compared to 21.6, 37.8, 29.0 and 34.5 for the RGB, cool, warm and neutral white treatments respectively. The monochromatic treatments (red, blue and green) were not included in the analysis due to the confounding effects of lack of other wavelengths. These treatments, however, were included on all Figures to provide references of response to monochromatic light.

To mitigate confounding factors for the effect of GL, statistical analysis only included the three treatments (RB, warm and RGB treatments) (Fig. 1) with comparable red and blue light to mitigate confounding factors (Table 1). The other treatments were included on all figures to provide a reference to the responses to these treatments.

Statistically significant differences for each parameter on a BL basis are between the four treatments (RGB, cool, warm and neutral white) (Fig. 1) with comparable red and green light (Table 1). Statistically significant differences for each parameter on a GL basis are between the three treatments (RB, warm and RGB treatments) (Fig. 1) with comparable red and blue light (Table 1).

All figures are arranged in order of general sensitivity to blue light with tomato being the most sensitive followed by cucumber and ending with wheat as the least sensitive species.

Dry mass

Effect of blue light

Dry mass (DM) significantly decreased as BL increased for tomato, cucumber, and pepper at the higher light level among comparable treatments (Fig. 4). Dry mass slightly decreased with increasing BL for soybean and wheat at the higher light level, but the effect was not statistically significant. Tomato was the only species at the lower light level that had a significant decrease in DM with increasing BL.

As expected, DM greatly increased with the two and a half fold increase in PPF. For tomato, radish, soybean, lettuce and wheat, DM was nearly two and half times greater at high light level than the lower light level, but for cucumber and pepper, DM was only 40 % greater at higher light level than the lower light level.

Overall the highest DM for all species at both light levels occurred in the treatments with 11-15 % BL and the effects of increasing BL were more pronounced at the higher light level.

Effect of green light

Dry mass significantly decreased with increasing GL only for radish at the higher light level among the comparable treatments (Fig. 5). There were no significant differences at the lower light level. For all other species there was minimal change in DM as GL increased from 0 to 30 % at either light level.

Dry mass greatly increased with PPF, but there were no consistent responses between PPF and GL.

Leaf area index*Effect of blue light*

Leaf area index (LAI) significantly decreased with increasing BL in tomato, cucumber, radish and pepper at the higher light level among comparable treatments (Fig. 6). At the lower light level, LAI significantly decreased with increasing BL only in tomato.

As expected, leaf area index increased with PPF. Similar to DM, LAI was typically highest in the treatments with 11-15 % BL for all species at both light levels.

Effect of green light

Leaf area index significantly increased with increasing GL for cucumber and wheat at the higher light level among the comparable treatments (Fig. 7). The only species at the lower light level that resulted in a significant decrease in LAI with increasing GL was lettuce. Leaf area index increased with PPF, except for pepper.

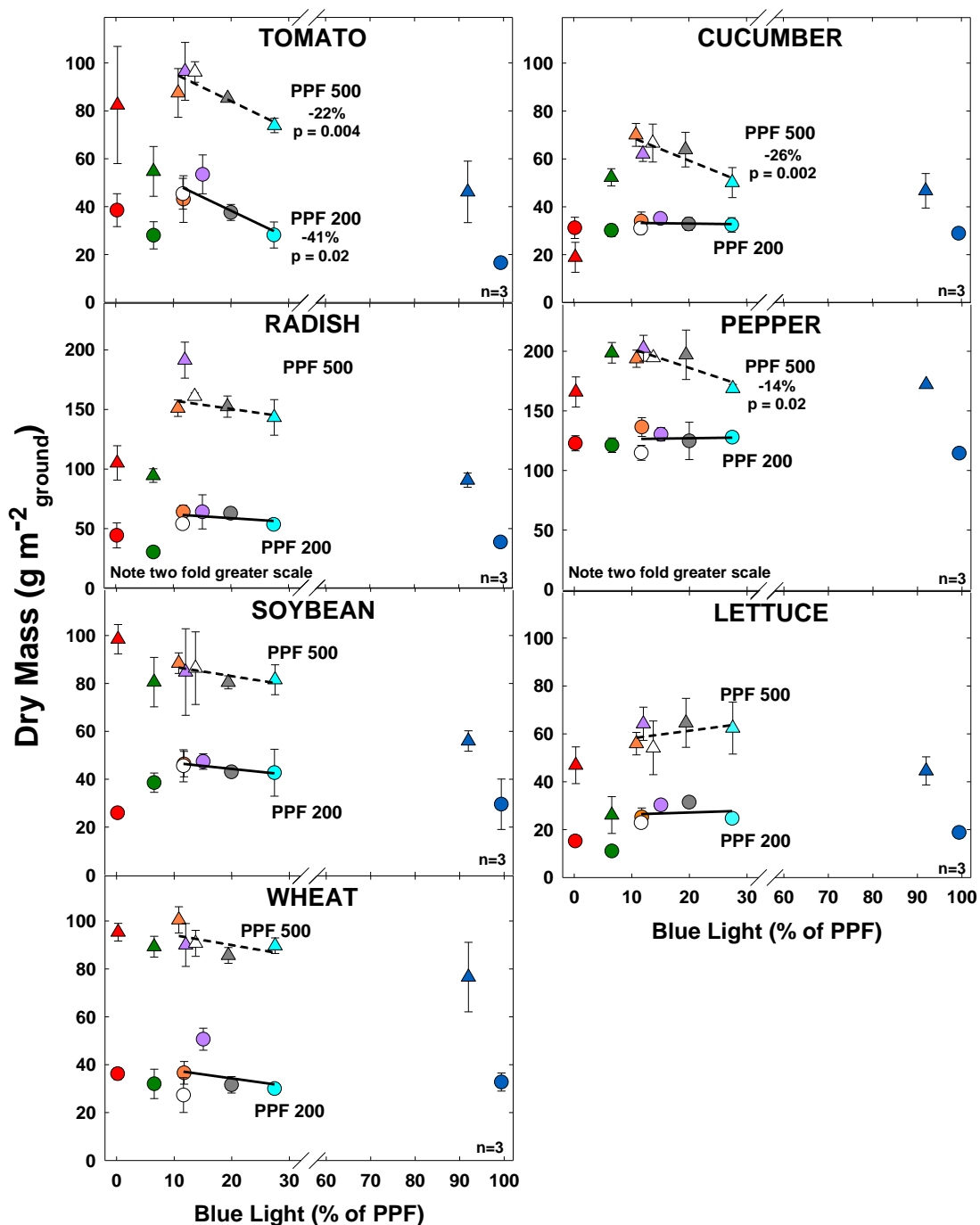


Fig. 4. The effect of percent blue light on dry mass (DM) for seven species under two PPFs. Note scale break for percent BL between 30 and 60. Also note two-fold scale increase for DM in radish and pepper. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the four treatments with comparable green and red wavelengths for each PPF. When significant, p-values and percent change are shown.

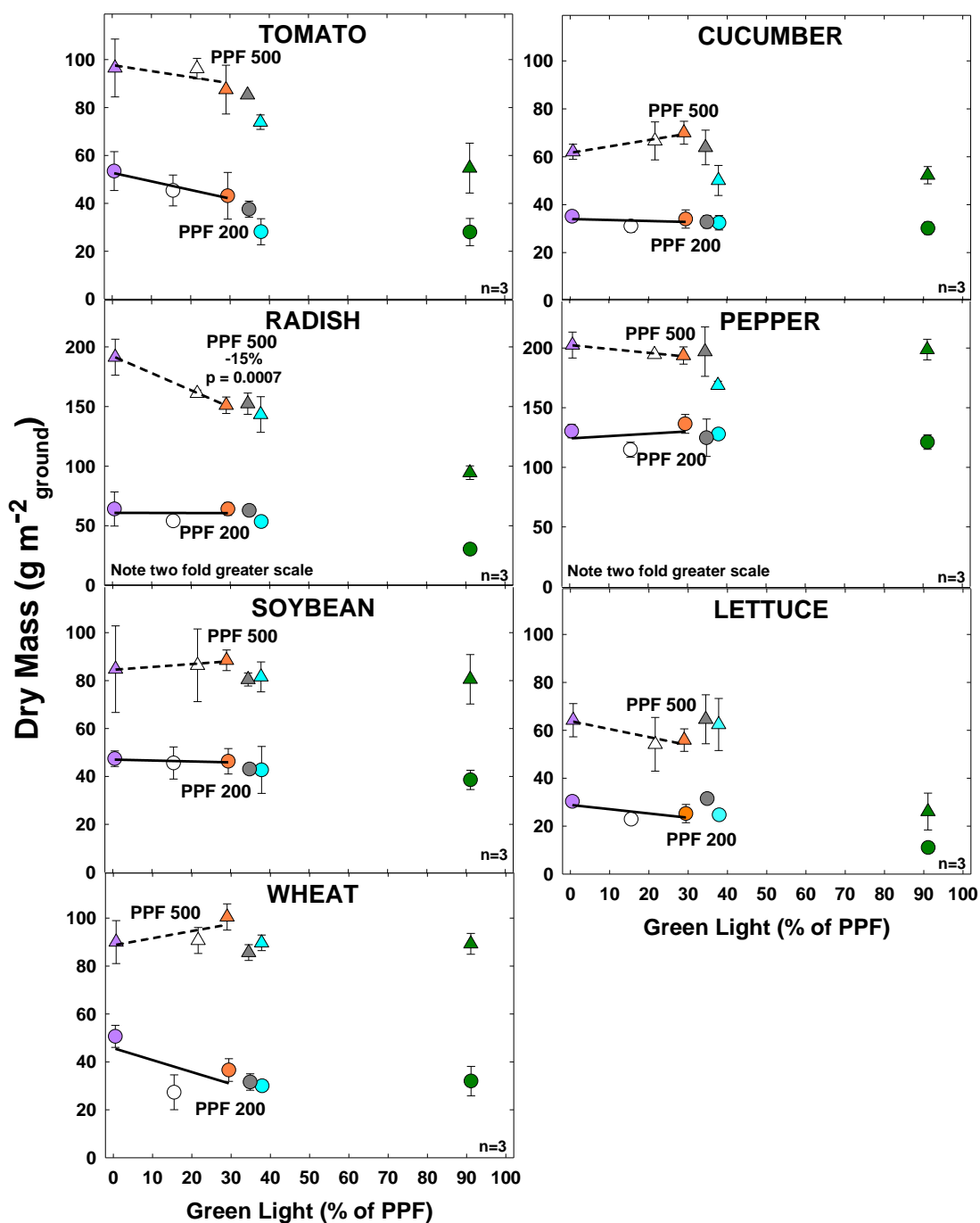


Fig. 5. The effect of percent green light on dry mass (DM) for seven species under two PPFs. Note two-fold scale increase for DM in radish and pepper. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the three treatments with comparable blue and red wavelengths for each PPF. When significant, p-values and percent change are shown.

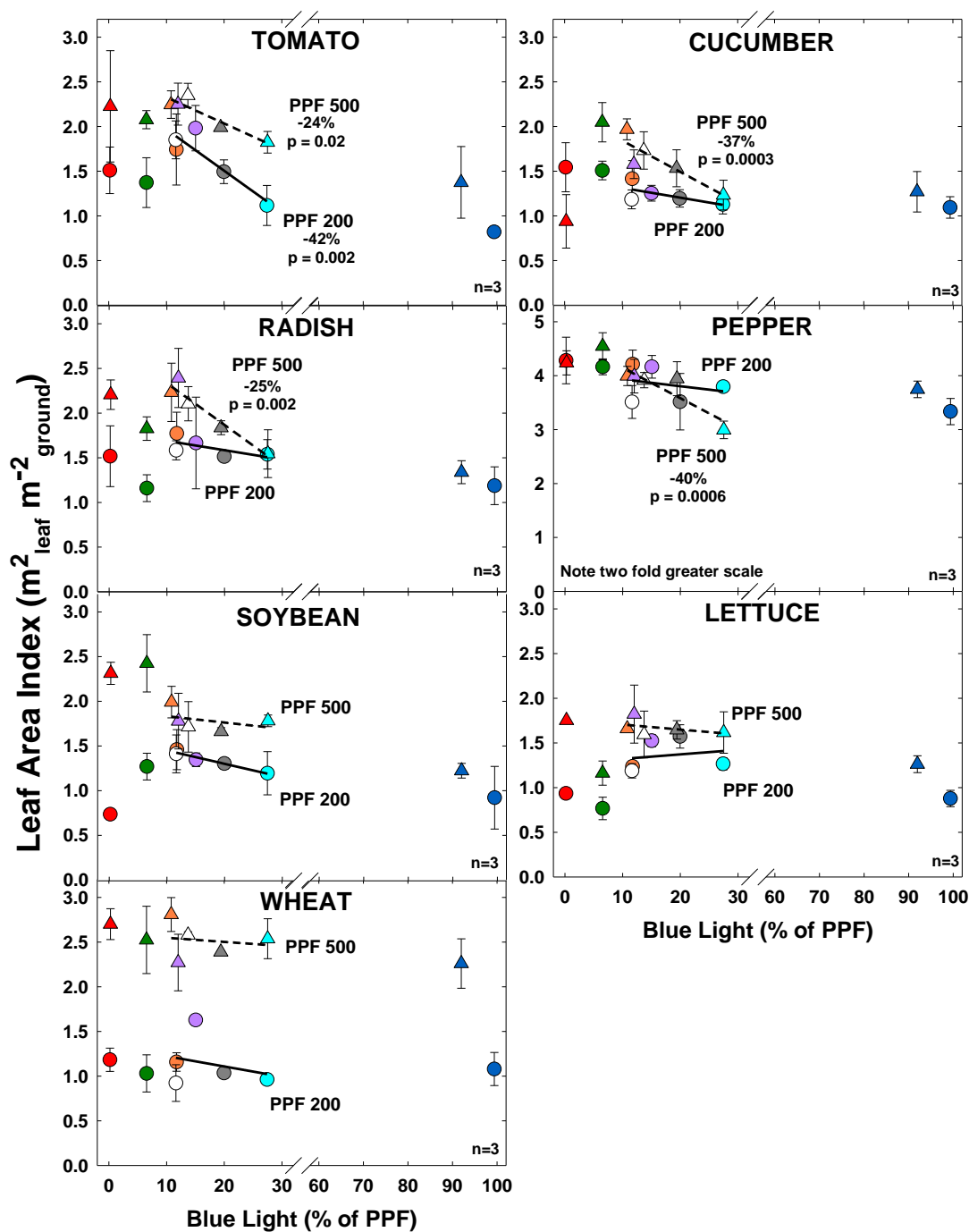


Fig. 6. The effect of percent blue light on leaf area index (LAI) for seven species under two PPFs. Note scale break for percent BL between 30 and 60. Also note two fold scale increase for pepper. Each data point shows the mean and standard deviation of three replicate studies for each species (n=3). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the four treatments with comparable green and red wavelengths for each PPF. When significant, p-values and percent change are shown.

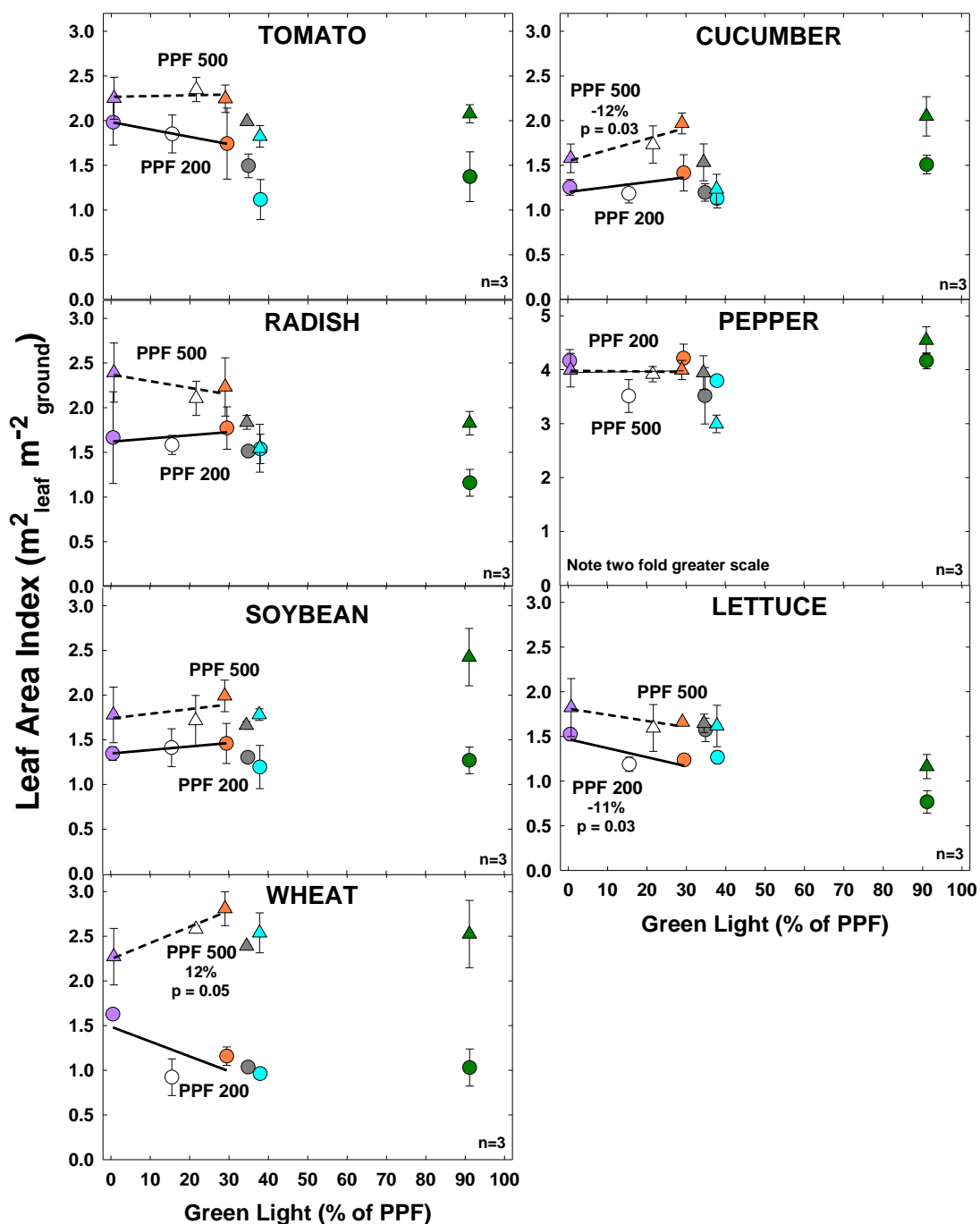


Fig. 7. The effect of percent green light on leaf area index (LAI) for seven species under two PPFs. Note two fold scale increase for pepper. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the three treatments with comparable blue and red wavelengths for each PPF. When significant, p -values and percent change are shown.

Net assimilation

Effect of blue light

Net assimilation significantly increased with increasing BL in cucumber, radish, pepper and lettuce at the higher light level among comparable treatments (Fig. 8). At the lower light level, net assimilation significantly increased with increasing BL only in cucumber among comparable treatments. For all other species there was minimal change in net assimilation as BL increased at either light level.

Net assimilation greatly increased with PPF for each species. Overall the highest net assimilation occurred in the treatments with 20-30 % BL for all species at both light levels.

Effect of green light

There were no significant differences in net assimilation with increasing GL at the higher light level among comparable treatments (Fig. 9). At the lower light level, net assimilation significantly decreased with increasing GL for cucumber and soybean among comparable treatments. For all other species there was minimal change in net assimilation as GL increased at the lower light level.

Net assimilation greatly increased with PPF for each species. Overall the highest net assimilation occurred in the treatments with the lowest GL for all species at both light levels, except pepper.

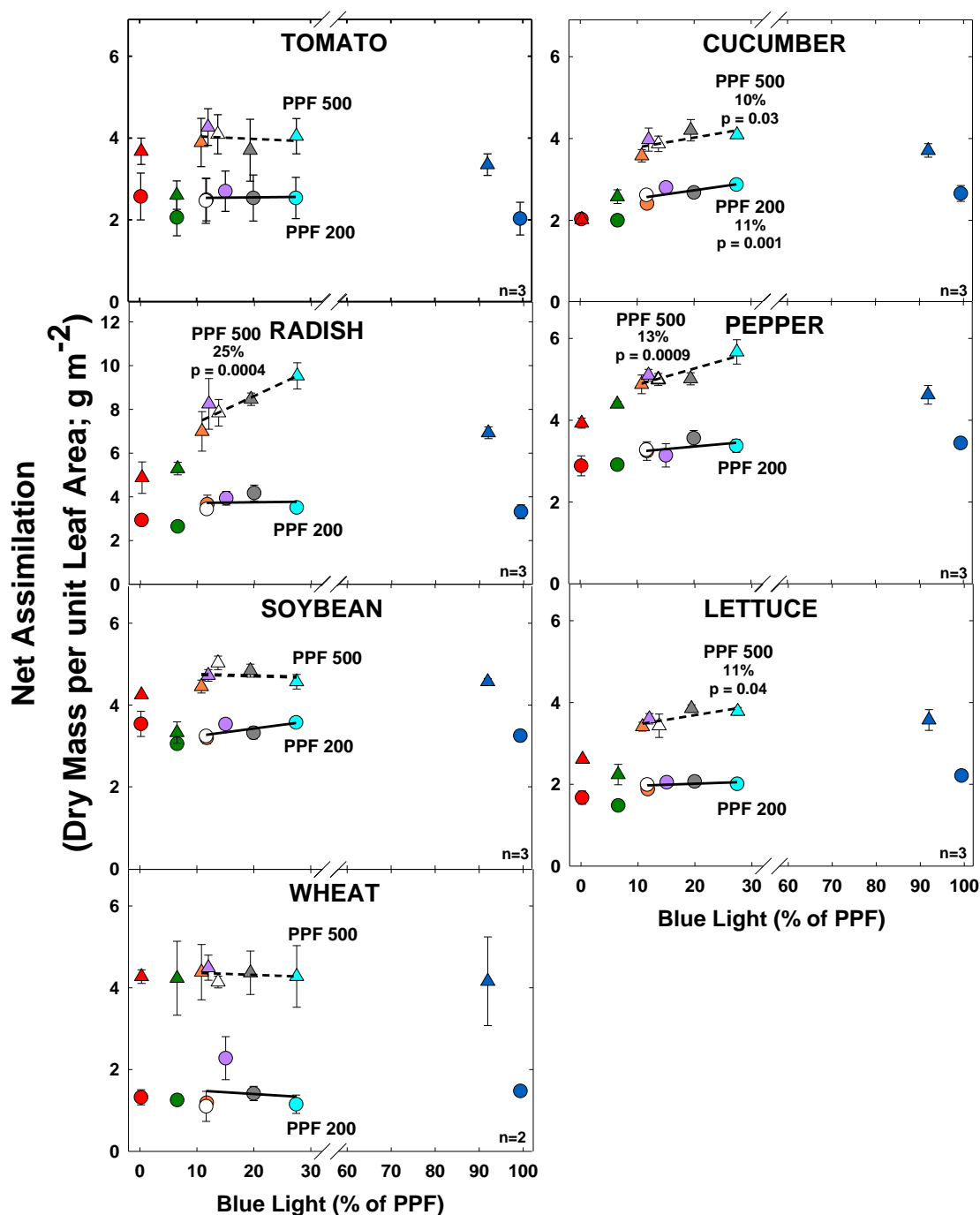


Fig. 8. The effect of percent blue light on net assimilation for seven species under two PPFs. Note scale break for percent BL between 30 and 60. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the four treatments with comparable green and red wavelengths for each PPF. When significant, p-values and percent change are shown.

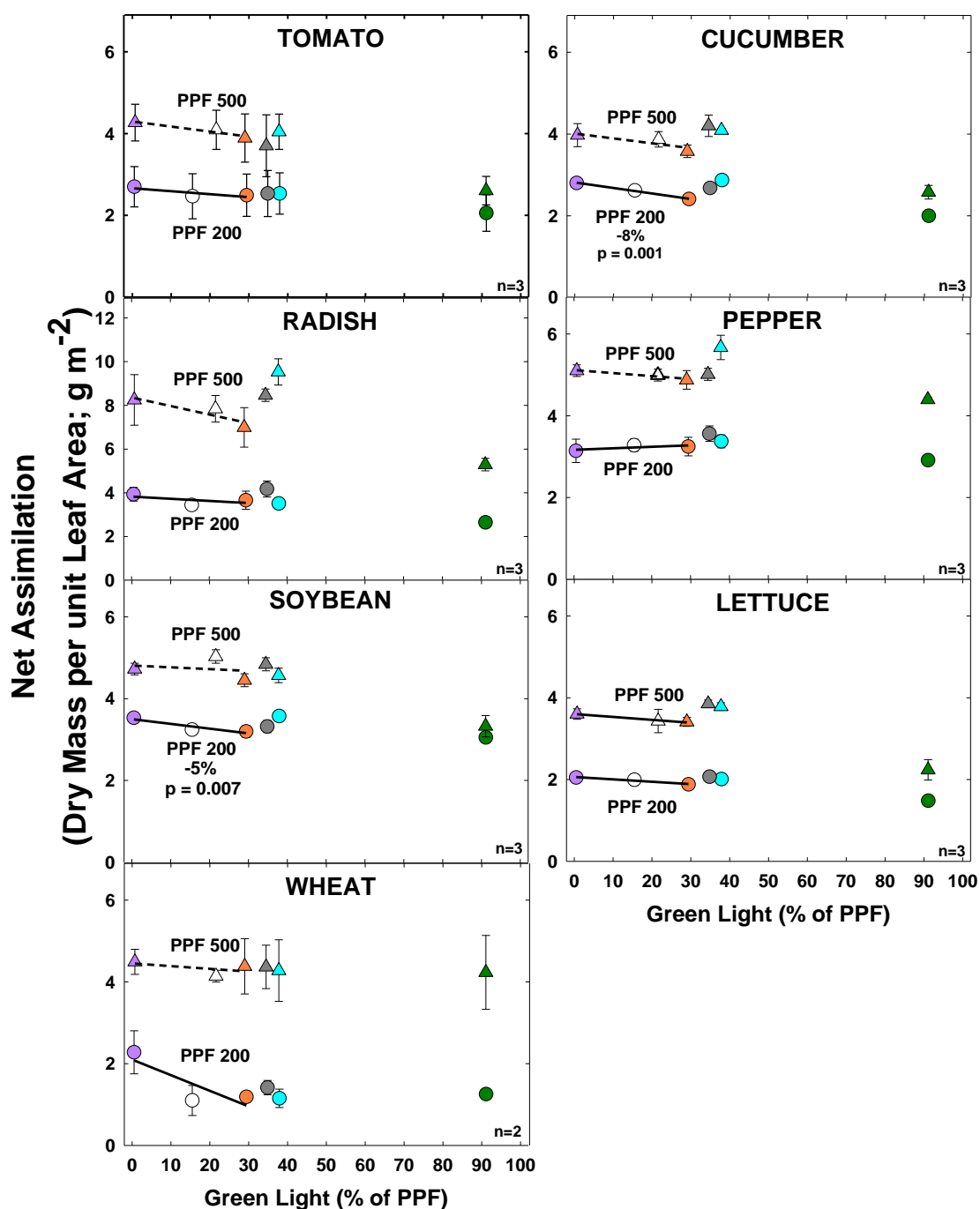


Fig. 9. The effect of percent green light on net assimilation for seven species under two PPFs. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the three treatments with comparable blue and red wavelengths for each PPF. When significant, p-values and percent change are shown.

Stem length

Effect of blue light

Stem length significantly decreased with increasing BL for tomato, cucumber, and pepper at the higher light level among comparable treatments (Fig. 10). Stem length was slightly decreased with increasing BL for radish, soybean, lettuce and wheat at the higher light level; however, results were not statistically significant. At the lower light level, stem length significantly decreased with increasing BL for tomato, pepper and soybean. For all other species at the lower light level there was minimal change in stem length as BL increased.

Stem length was similar between PPF levels except for pepper, soybean and wheat. Overall the highest stem length for all species at both light levels, except pepper and wheat, typically occurred in the treatments in the lower BL treatments.

Effect of green light

Stem length significantly increased with increasing GL only for tomato at the higher light level among the comparable treatments (Fig. 11). For all other species there was minimal change in stem length as GL increased at the higher light level. At the lower light level, stem length significantly increased with increasing GL for pepper, soybean and lettuce. For all other species at the lower light level there was minimal change in stem length as GL increased.

Stem length was similar between PPF levels for all species except soybean and wheat. Overall the highest stem length typically occurred in the treatments with lower GL fraction for all species at both light levels.

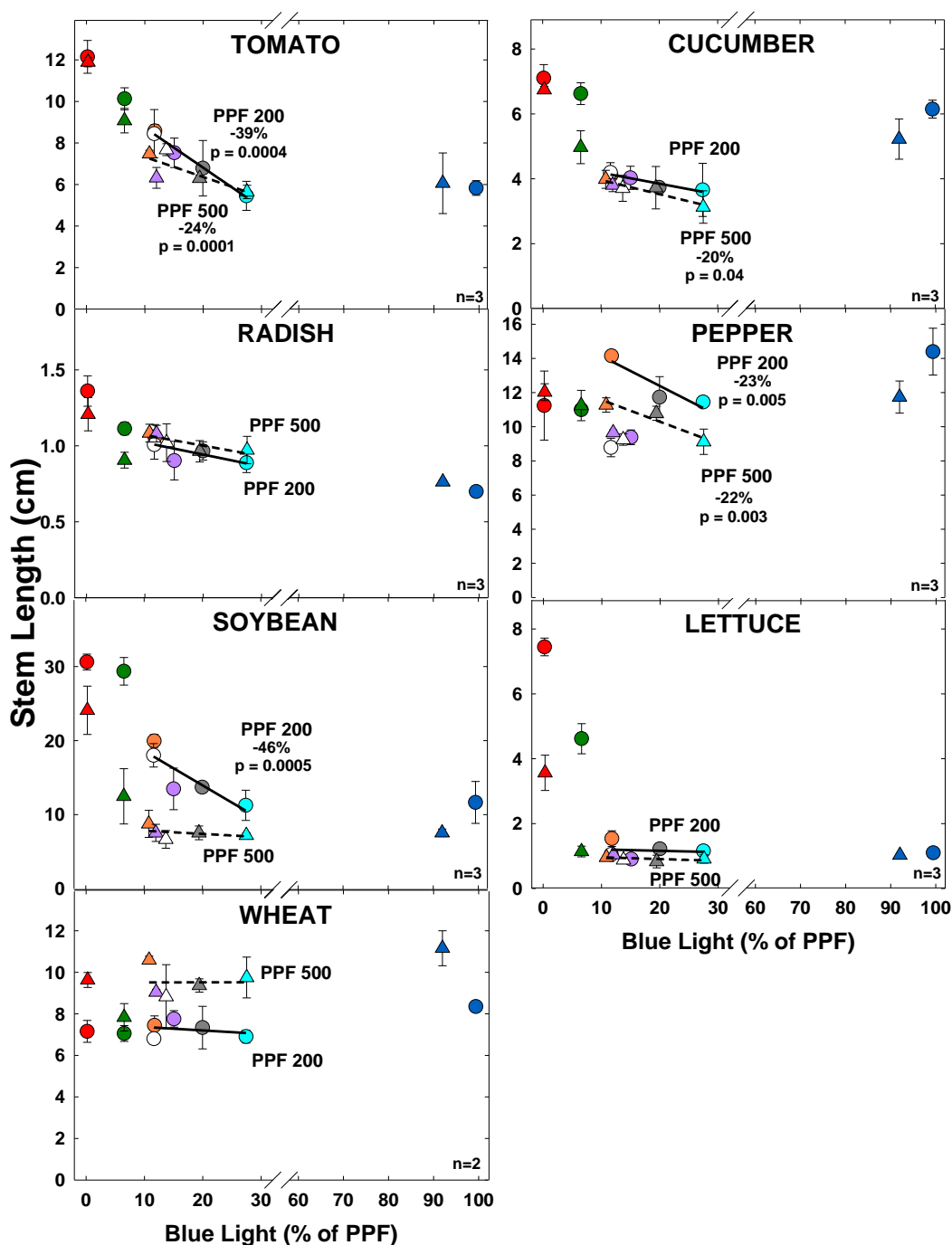


Fig. 10. The effect of percent blue light on stem length for seven species under two PPFs. Note scale break for percent BL between 30 and 60. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the four treatments with comparable green and red wavelengths for each PPF. When significant, p-values and percent change are shown.

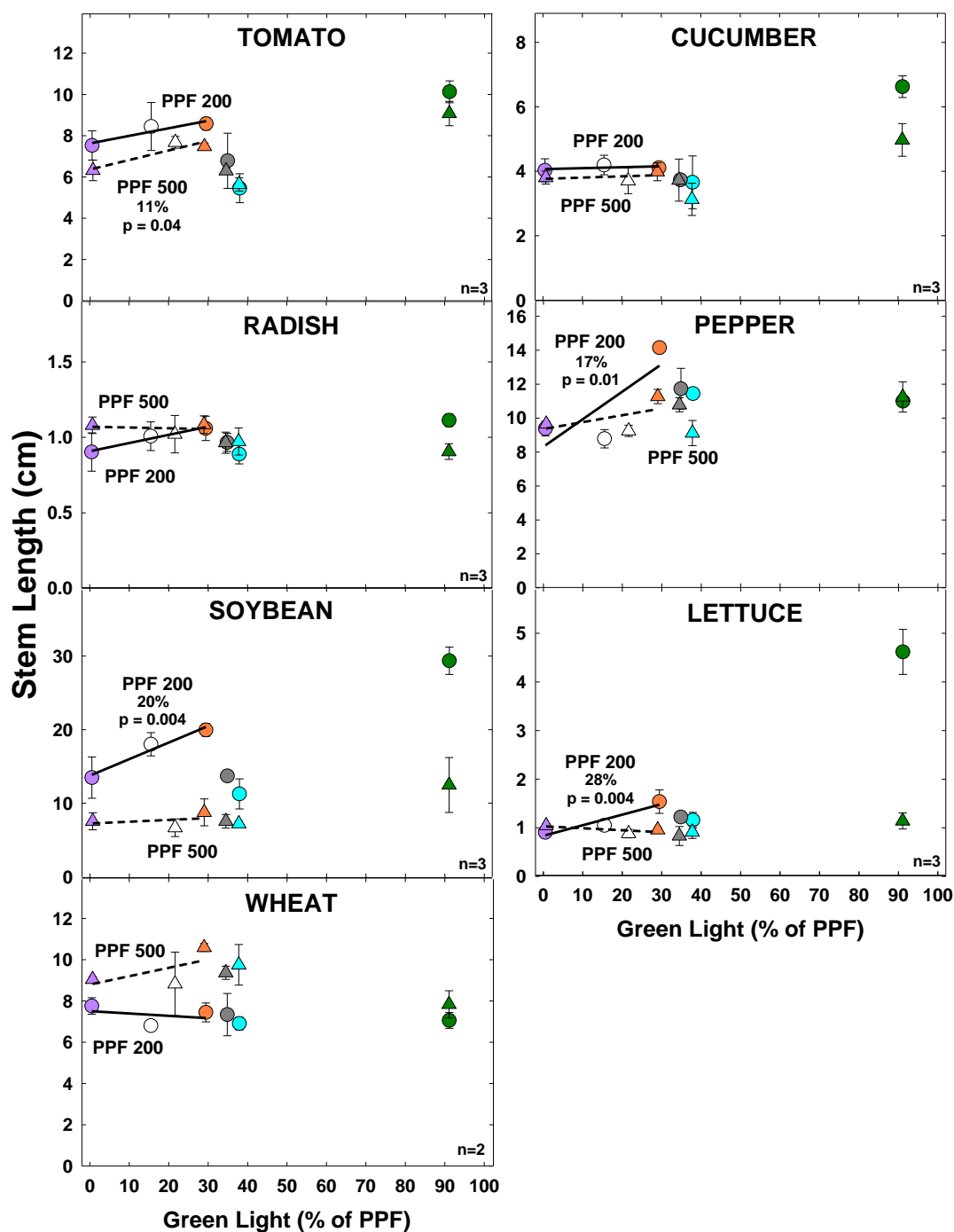


Fig. 11. The effect of percent green light on stem length for seven species under two PPFs. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the three treatments with comparable blue and red wavelengths for each PPF. When significant, p-values and percent change are shown.

Petiole length

Effect of blue light

For these results lettuce and wheat are not included due to these species not producing petioles as a major form of their development. Petiole length significantly decreased with increasing BL for tomato, cucumber and radish at the higher light level among comparable treatments (Fig. 12). For all other species at the higher light level there was minimal change in petiole length as BL increased. At the lower light level, petiole length significantly decreased with increasing BL only for cucumber. For all other species at the lower light level petiole length slightly decreased as BL increased.

Petiole length decreased as PPF increased for all species except cucumber. Similar to specific leaf area, this parameter produced higher petiole length at the lower light as a way to increase capture of incident radiation. Overall the highest petiole length was variable between species but generally occurred in the treatments that contained the least BL for all species at both light levels, except cucumber.

Effect of green light

For these results lettuce and wheat are not included due to these species not producing petioles as a major form of their growth. Petiole length significantly increased with increasing GL only for radish at the higher light level among comparable treatments (Fig. 13). For all other species at the higher light level there was minimal change in petiole length as GL increased. At the lower light level, petiole length significantly increased with increasing GL for cucumber, pepper and soybean. For all other species at the lower light level there was minimal change in petiole length as GL increased.

Petiole length decreased as PPF increased for each species. Similar to specific leaf area, this parameter produced higher petiole length at the lower light as a way to increase capture of incident radiation. Overall the highest petiole length generally occurred in the treatments with the most GL for all species at both light levels.

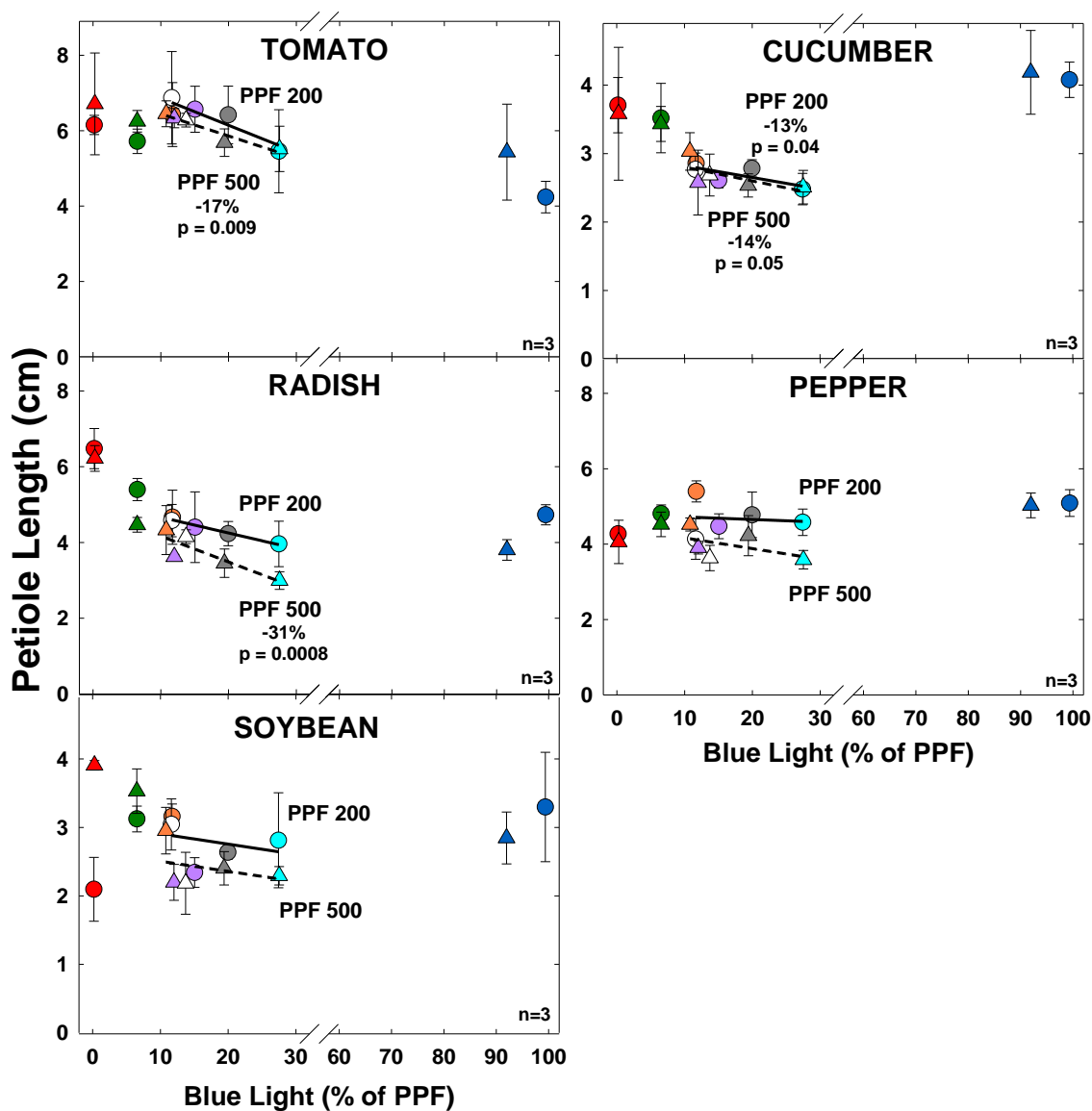


Fig. 12. The effect of percent blue light on petiole length for seven species under two PPFs. Note scale break for percent BL between 30 and 60. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the four treatments with comparable green and red wavelengths for each PPF. When significant, p-values and percent change are shown.

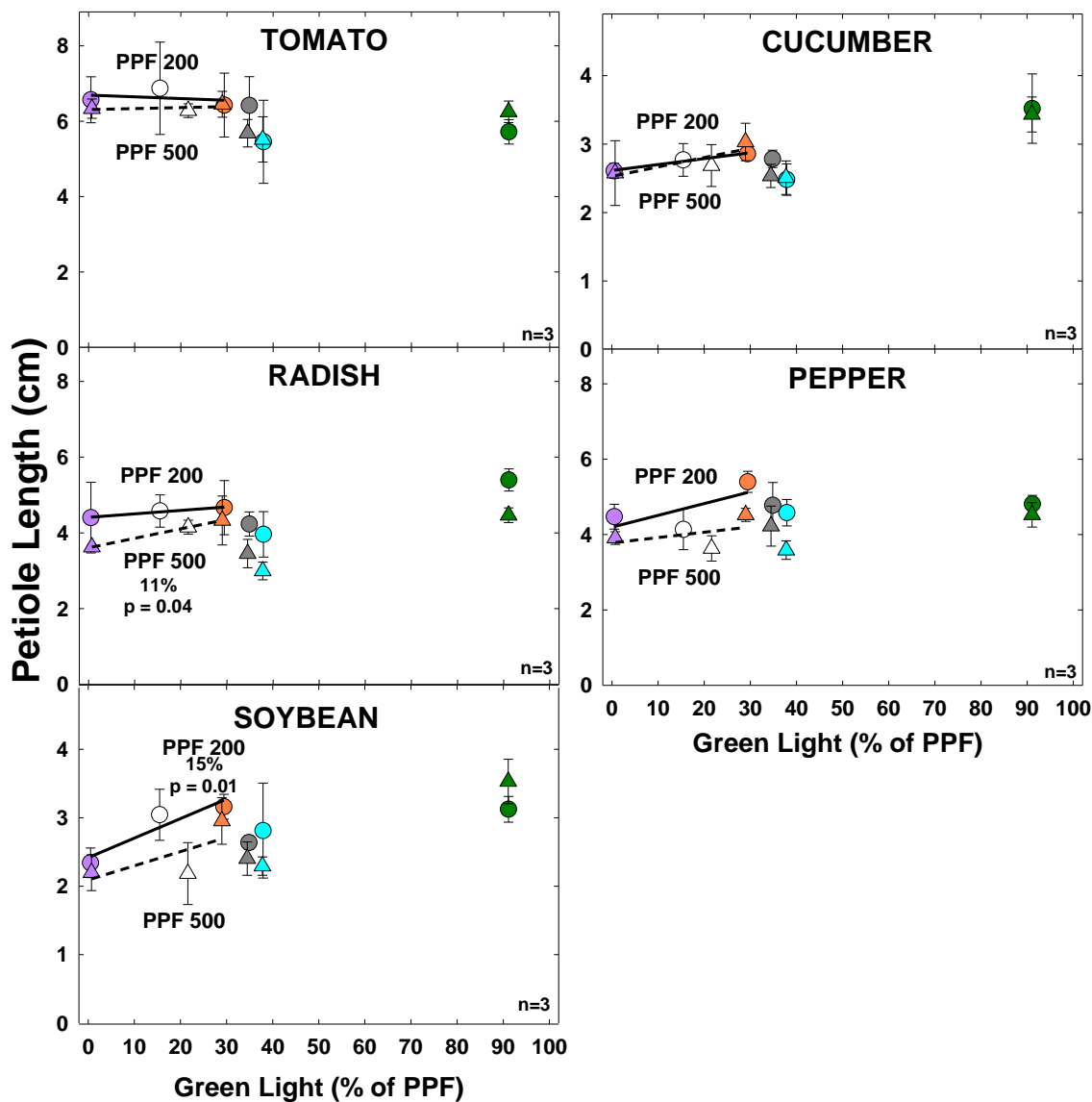


Fig. 13. The effect of percent green light on petiole length for seven species under two PPFs. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the three treatments with comparable blue and red wavelengths for each PPF. When significant, p -values and percent change are shown.

Specific leaf area

Effect of blue light

Specific leaf area significantly decreased with increasing BL for radish, pepper and lettuce at the higher light level among comparable treatments (Fig. 14). For all other species at the higher light level there was minimal change in specific leaf area as BL increased. At the lower light level, specific leaf area significantly decreased with increasing BL only for cucumber. For all other species at the lower light level there was minimal change in specific leaf area as BL increased.

Specific leaf area greatly decreased as PPF increased for each species. This occurred as a result of leaves being thicker at the lower light level due to decreased intensity and slower growth. Overall the highest specific leaf area occurred in treatments with the least BL for all species at both light levels.

Effect of green light

Specific leaf area significantly increased with increasing GL only for pepper at the higher light level among the comparable treatments (Fig. 15). For all other species there was minimal change in specific leaf area as GL increased at the higher light level. At the lower light level, specific leaf area significantly increased with increasing GL for tomato, cucumber and soybean. For all other species at the lower light level there was minimal change in stem length as GL increased.

Specific leaf area greatly decreased as PPF increased for each species. This occurred as a result of leaves being thicker at the lower light level due to decreased

intensity and slower growth. Overall the highest specific leaf area typically occurred in treatments with the most GL for all species at both light levels, except wheat.

Chlorophyll

Effect of blue light

Chlorophyll concentration significantly increased with increasing BL in tomato, cucumber, radish and pepper at the higher light level among comparable treatments (Fig. 16). There was minimal change in chlorophyll concentration as BL increased for all other species at the higher light level. At the lower light level, chlorophyll concentration significantly increased with increasing BL only for tomato. For all other species at the lower light level, there was minimal change in chlorophyll concentration as BL increased.

Chlorophyll concentration increased with PPF. Overall the highest chlorophyll concentration at both light levels typically occurred in the treatments with 20-30 % BL.

Effect of green light

Chlorophyll concentration significantly decreased with increasing GL only in cucumber at the higher light level among the comparable treatments (Fig. 17). At the lower light level, chlorophyll concentration significantly decreased with increasing GL in tomato, cucumber, pepper and lettuce among the comparable treatments. For all other species at the lower light level there was minimal change in chlorophyll concentration and GL increased. Chlorophyll concentration increased with PPF for each species; however, responses were not consistent between PPF and GL.

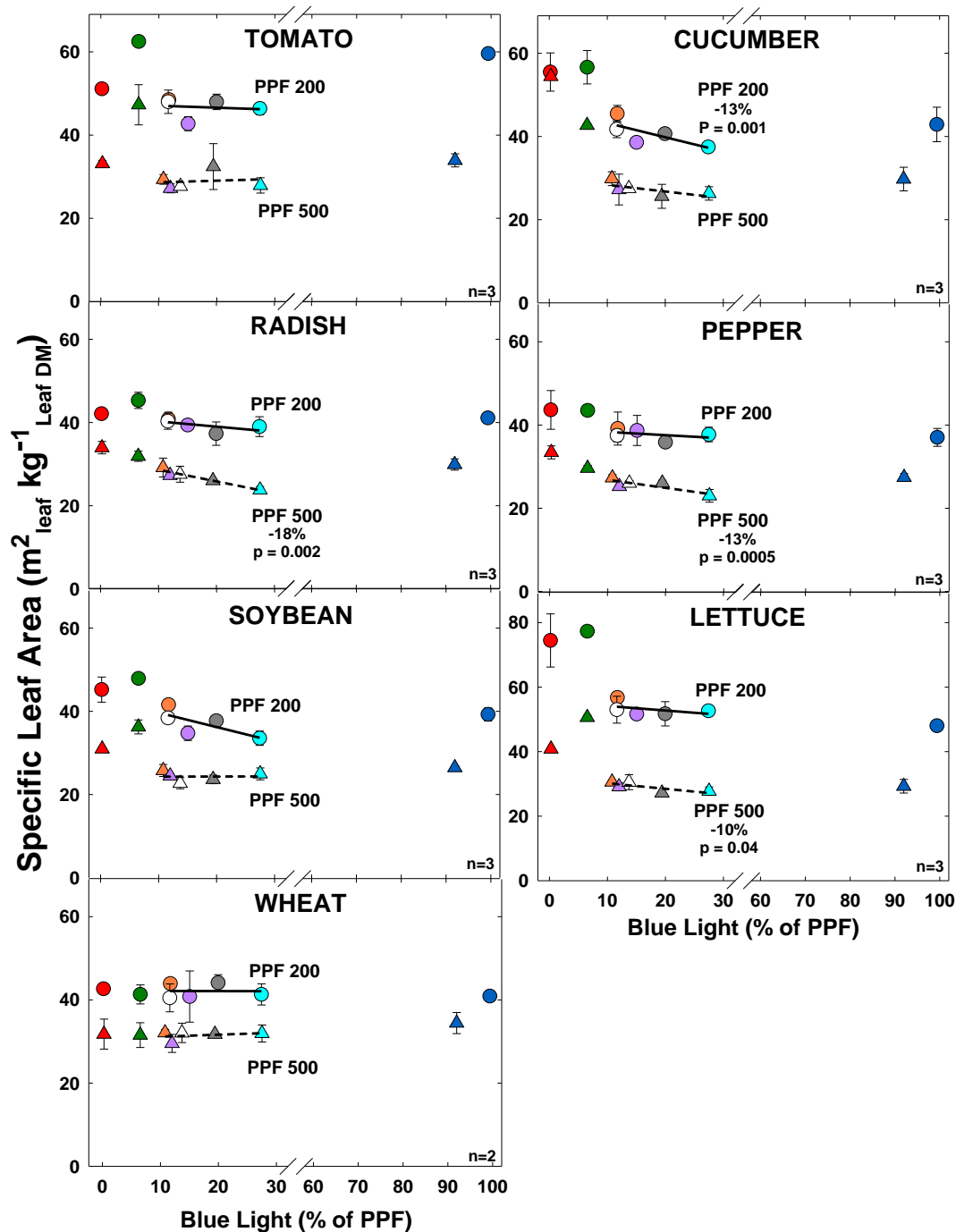


Fig. 14. The effect of percent blue light on specific leaf area for seven species under two PPFs. Note scale break for percent BL between 30 and 60. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the four treatments with comparable green and red wavelengths for each PPF. When significant, p-values and percent change are shown.

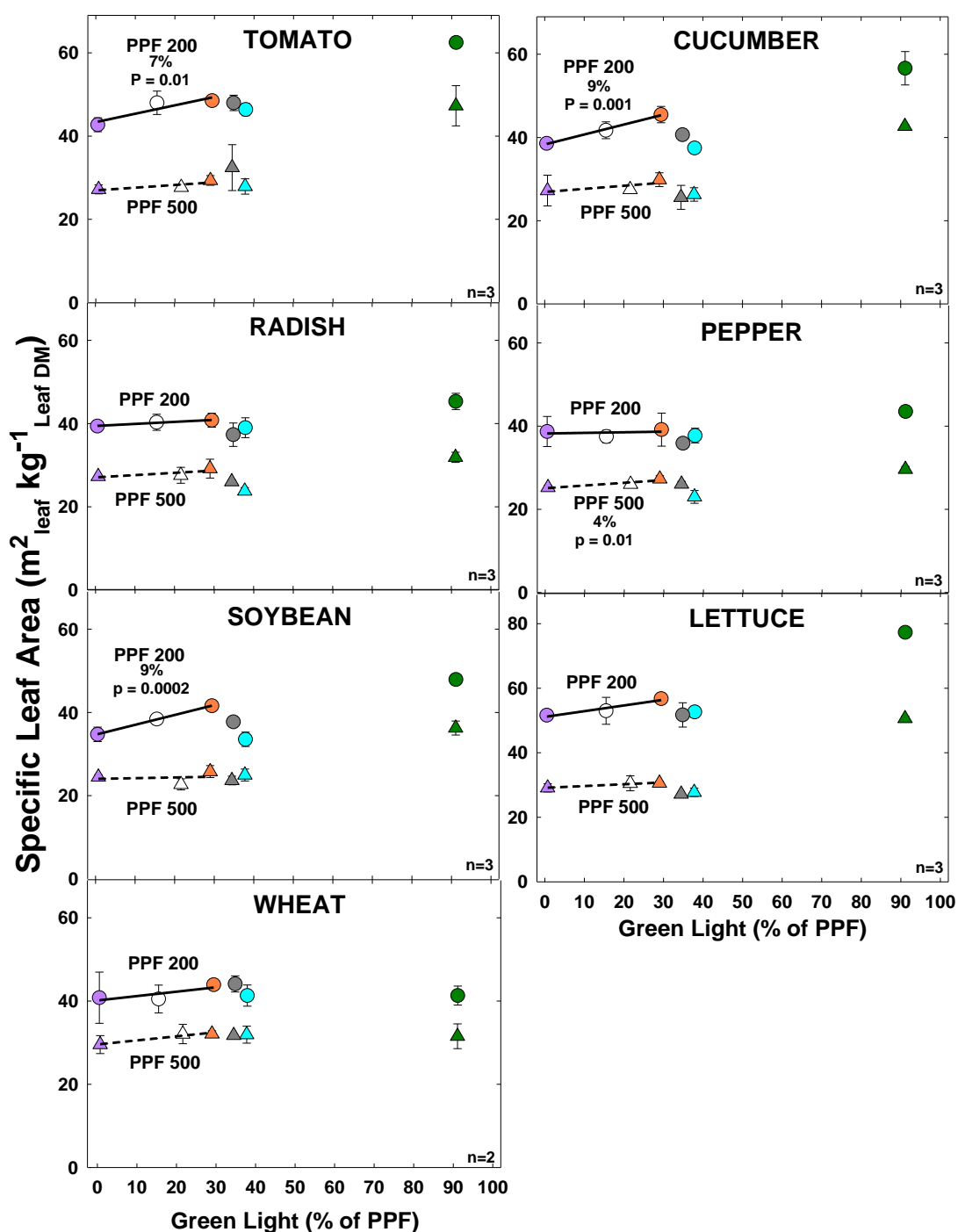


Fig. 15. The effect of percent green light on specific leaf area for seven species under two PPFs. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the three treatments with comparable blue and red wavelengths for each PPF. When significant, p-values and percent change are shown.

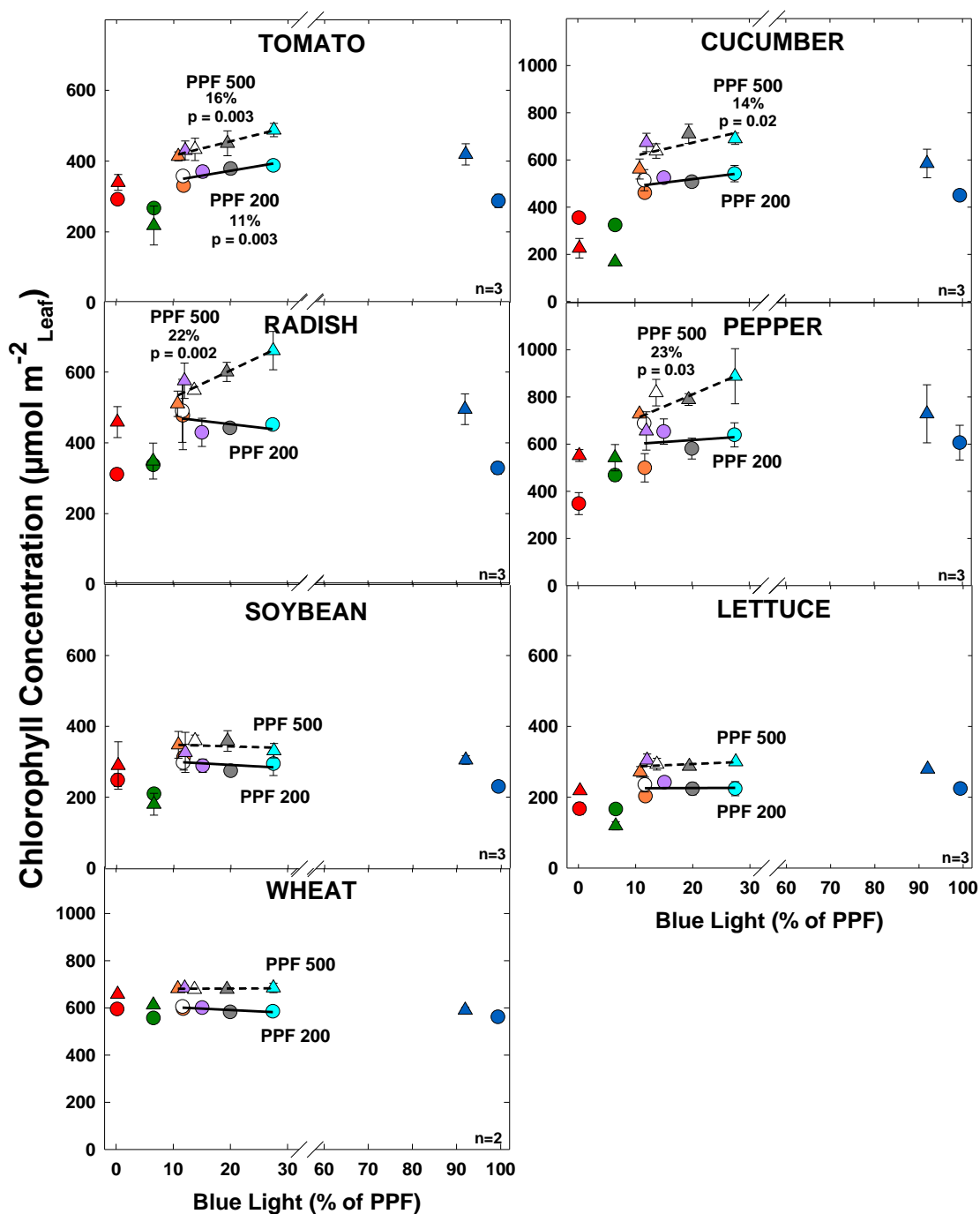


Fig. 16. The effect of percent blue light on chlorophyll concentration for seven species under two PPFs. Note scale break for percent BL between 30 and 60. Each data point shows the mean and standard deviation of three replicate studies for each species (n=3). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the four treatments with comparable green and red wavelengths for each PPF. When significant, p-values and percent change are shown.

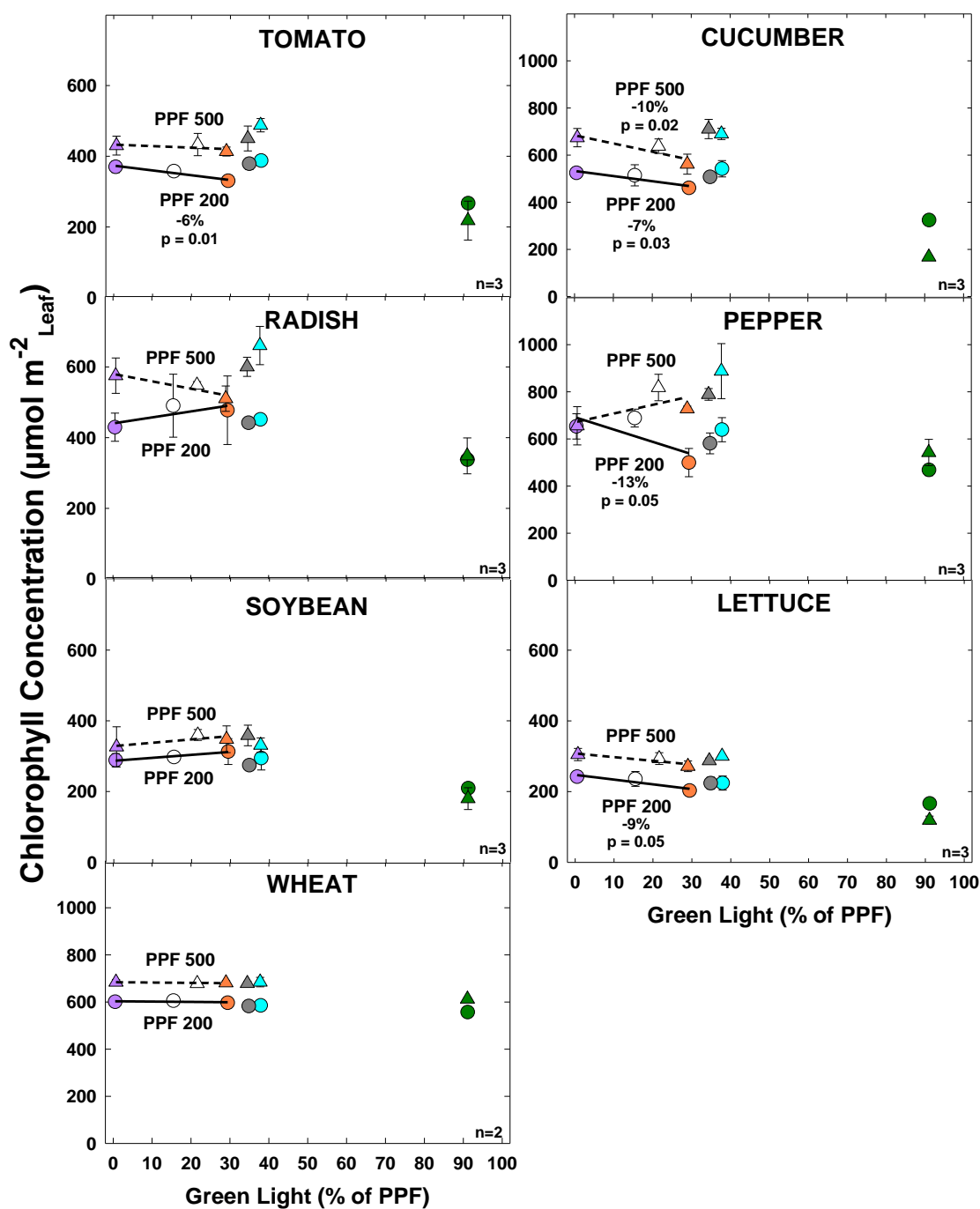


Fig. 17. The effect of percent green light on chlorophyll concentration for seven species under two PPFs. Each data point shows the mean and standard deviation of three replicate studies for each species ($n=3$). Some error bars are smaller than the symbol size. See Fig. 1 and Table 1 for symbol color and shape legend. To minimize confounding spectral effects the regression line includes only the three treatments with comparable blue and red wavelengths for each PPF. When significant, p-values and percent change are shown.

DISCUSSION

Definition of growth and development

Plant growth can be defined as DM produced and is the combined result of LAI and net assimilation (Hunt, 1982; Leopold and Kriedemann, 1975). Plant development can be defined as leaf expansion, stem elongation, and petiole length which determine LAI and light interception. Plants adapt to their light environment to maximize light interception and growth (Hogewoning et al., 2012). This includes modification of stem and petiole length, leaf thickness, cell size and number in leaf and stem tissue, and cell arrangement and function (Dougher and Bugbee, 2004; Schuerger et al., 1997; Wang et al., 2011).

Effects of blue light

The highest DM, greatest LAI and longest stem length typically occurred in the lowest BL treatments at both PPFs, where plants tended to exhibit shade-avoidance responses. As BL increased cryptochrome may become overstimulated and contribute to significantly reduced LAI, which would decrease radiation capture and ultimately growth.

Our results are similar to Hernández and Kubota (2015) for cucumber in which both leaf area and dry mass decreased with increasing BL over a similar range of BL fraction. Hernández and Kubota (2015) also indicate that leaf area and dry mass continue to decrease up to 75 % BL. Results of the current study for wheat, soybean and lettuce are similar to those of Dougher and Bugbee (2001) for dry mass, leaf area, stem length, chlorophyll, specific leaf at the same two light levels and across a similar BL range using

filtered metal halide and high pressure sodium lamps. Dougher and Bugbee (2001) concluded that BL had only a small effect on the plant morphology for wheat, and indicated that the response may be associated with the erectophile morphology of a monocot. Our results are also similar to Cope et al. (2014) for lettuce, radish and pepper at the same two light levels and across similar parameters for growth and development. However, Cope et al. (2014) analyzed the effects of BL for each species across all BL treatments (0 to 92 % BL) which included confounding factors potentially leading to conclusions that are associated with other variables.

Contrary to our results for radish, Yorio et al. (2001) concluded that their RB (10 % BL) treatment had significantly lower growth compared to cool white fluorescent (16 % BL) for radish; however, lettuce growth was similar between the two light sources. The cool white fluorescent treatment had nearly 47 % more GL compared to the RB treatment, so differences in growth cannot be entirely attributed to BL.

Hoenecke et al. (1992) grew lettuce seedlings for six days under multiple BL treatments ranging from 0 to 40 % BL at two PPFs (150 and 300 $\mu\text{mol m}^{-2} \text{s}^{-1}$). Similar to our results for lettuce stem length, they reported that hypocotyl length rapidly decreased as BL increased up to 28 %. Their results are also consistent with those of Dougher and Bugbee (2001), although total stem length was reported, not hypocotyl length. Similar to our results, Brown et al. (1995) reported that pepper stem length decreased as BL increased up to 21 % BL.

The current study used a classic method of determining net assimilation (photosynthetic efficiency) using crop growth analysis and leaf area index as described in

Leopold and Kriedemann (1975) and Hunt (1982). The ratio of growth to leaf area index provides a measure of net assimilation integrated over time. Because photon flux was constant at either 200 or 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ this is a measure of photosynthetic efficiency. Poorter and Remkes (1990) studied growth rate in 24 wild species and concluded that net assimilation rate did not correlate well with dry mass production, however LAI did have a high correlation with dry mass production. This result is evident in DM, LAI and net assimilation results of the current study in which DM and LAI decreased at similar rates with increasing BL compared to net assimilation which had no change or increased slightly with increasing BL. Goins et al. (2001) studied the effects of different wavelengths of RL and a constant BL level (8-9 % BL) from LEDs on lettuce and radish and concluded that increased incident radiation capture resulted in increased growth more than higher individual leaf photosynthetic rates. Our study answers a question identified by Goins et al. (2001) that continuing to increase BL continues to decrease LAI, therefore radiation capture. Hogewoning et al. (2010b) also found similar effects in cucumber grown under artificial solar, fluorescent and high pressure sodium lamps. Growth of cucumber was at least one and a half times greater under the artificial solar treatment, which was related to more efficient light interception with no change in photosynthesis.

Goins et al. (1997) and Yorio et al. (2001) demonstrated that some blue light was necessary to improve photosynthetic efficiency. Hogewoning et al. (2010a) found that photosynthetic capacity in high light continued to increase with increasing BL up to 50 % BL, which is similar to our study, except that our BL range had a maximum of 28 % BL. Hogewoning et al. (2010a), however, used a PPF of 100, which is significantly lower

than the current study. Our results were also similar to Ouzounis et al. (2015) for lettuce in which no differences were seen in photosynthetic efficiency at the lower light level, however our results produced a significant increase in photosynthetic efficiency at the higher light level, which indicate an interaction between light quality and PPF for lettuce. Also similar to our study, Terfa et al. (2013) showed that increasing blue light from 5 to 20 % increased leaf thickness (specific leaf area) and increased photosynthetic capacity. Hernández and Kubota (2015) also found that net photosynthesis increased in cucumber as blue light fraction increased from 10 to 80 %.

Effects of green light

Green light can alter plant development (Folta and Maruhnich, 2007), but our results were inconsistent between light levels and species. Also our results contrast to findings of Wang and Folta (2013) that effects may decrease as PPF increases. However, duration of trials in the current study was for 21 days, which was prior to full canopy closure and the contribution of GL may increase as canopy closure occurs.

The highest dry mass tended to be the lowest green light treatment but the effect was only statistically significant in radish at high light. Increasing GL had a minimal effect at the lower light level. Similarly, Johkan et al. (2012) reported that, in general, the green treatments were closer in total DM for lettuce to the cool white fluorescent treatment at 100 PPF, than at 200 and 300 PPF. Lin et al. (2013) reported that lettuce grown at 210 PPF under RB LEDs had a lower DM than lettuce grown under two broad spectrum light sources (red + blue + white LEDs and fluorescent lamps) at the same PPF. This is in contrast to findings of our study in which there was higher DM in the RB

treatment and DM showed no change or decreased under broad spectrum treatments (RGB, warm, neutral and cool white), which had increased GL compared to the RB treatment. Our results differ from findings for lettuce in Kim et al. (2004b) and for radish in Yorio et al. (2001), but agree with the results for lettuce in Yorio et al. (2001). However, when considering the interaction of PPF and light quality, as in Johkan et al. (2012), our results are similar with both Kim et al. (2004b) and Yorio et al. (2001). Kim et al. (2004b) reported that supplementing red and blue LEDs with green light (from green fluorescent lamps) increased lettuce growth by up to 48 % at the same total PPF. Their results indicated that too much (51 %) or too little (0 %) green light caused a decrease in growth, while about 24 % was optimal. This finding contradicts our results in which growth response was inconsistent as GL was added. The RB treatment (0 % GL) typically produced the highest growth for most species tested. Similar to our study, Hernández and Kubota (2015) concluded that GL (28 %) had no effect on cucumber growth.

Paradiso et al. (2011) measured photosynthesis of individual rose leaves at 18 wavelengths and using a deviation of the Beer-Lambert equation to calculate photosynthesis of a plant community with a leaf area index of three (LAI=3). Their results indicate that there is an increased utilization of GL in whole plant communities compared to individual leaves. These results, and those of the current study, suggest that photosynthetic efficiency at the individual leaf level (e.g. Sun et al. (1998) and Terashima et al. (2009)) should not automatically be extrapolated to the whole plant community level.

CONCLUSIONS

This was a comprehensive study with seven species and the effect of blue and green light fractions at PPFs of 200 and 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$. It is clear that plant growth and development was affected by interactions between PPF, light quality, plant species.

At a PPF of 500, increasing blue light from 11 to 28 % decreased dry mass in tomato, cucumber, radish, and pepper, but there was no significant effect on soybean, lettuce and wheat. At a PPF of 200, dry mass significantly decreased only in tomato across the blue light range. Effects on leaf area paralleled effects on dry mass in all species at both PPFs, indicating that the effects of blue light on dry mass were mediated by changes in leaf area.

In contrast to the significant effect of blue light dry mass and leaf area, increasing green light fraction from zero to 30 % resulted in few significant differences, and there was no consistent direction of the effect among species or PPF levels on DM, LAI or net assimilation. These results indicate that GL had little effect during initial growth stages, but its importance may increase over time as a dense canopy forms.

Historically, studies to understand spectral effects on plant growth have focused on single leaf photosynthetic efficiency over short time intervals. The results of current study contrast spectral efficiency curves of Hoover (1937), McCree (1972) and Inada (1976), which indicate the blue light is used less efficiently in photosynthesis. Therefore, it is apparent that other interacting factors alter the effect of light quality on photosynthetic efficiency in long-term studies.

This study used the classical technique of crop growth analysis to determine net assimilation (photosynthetic efficiency). There was no evidence of decreasing net assimilation, in any of the seven species, with increasing blue light. Leaf area index, however was significantly decreased for these species and suggest that the effect of blue light on reducing leaf area and radiation interception was the underlying cause of the reduction in growth rather than net assimilation. Since LAI determines radiation capture and is highly correlated with dry mass gain, it is apparent that improvements in radiation capture efficiency are responsible for nearly all of the increases in dry mass, and this usually resulted in decreased photosynthetic rate because of increased self-shading.

Collectively, these results indicate significant differences among species in sensitivity to blue light and less sensitivity to green light among species, and that the effect of blue light on dry mass is primarily determined by changes in leaf area.

REFERENCES

- Barta D, Tibbitts T, Bula R, Morrow R. Evaluation of light emitting diode characteristics for a space-based plant irradiation source. *Adv Space Res* 1992;12:141-49.
- Briggs WR, Olney MA. Photoreceptors in plant photomorphogenesis to date. Five phytochromes, two cryptochromes, one phototropin, and one superchrome. *Plant Phys* 2001;125:85-88.
- Broadersen CR, Vogelmann TC. Do changes in light direction affect absorption profiles in leaves? *Funct Plant Biol* 2010;37:403-12.
- Brown CS, Schuerger A, Sager JC. Growth and photomorphogenesis of pepper plants under red LEDs with supplemental blue or far-red lighting. *J Amer Soc Hort Sci* 1995;120:808-13.
- Bula R, Morrow R, Tibbitts T, Barta D, Ignatius R, Martin T. Light-emitting diodes as a radiation source for plants. *HortSci* 1991;26:203-05.
- Bullock D, Nielsen R, Nyquist W. A growth analysis comparison of corn grown in conventional and equidistant plant spacing. *Crop Sci* 1988;28:254-58.
- Casal JJ. Phytochromes, cryptochromes, phototropin: Photoreceptor interactions in plants *Photochem Photobiol* 2000;71:1-11.
- Chaves I, Pokorney R, Byrdin M, Hoang M, Ritz R, Brettel K, et al. The crytochromes: Blue light photoreceptors in plants and animals. *Annu Rev Plant Biol* 2011;62:335-64.
- Chen X-l, Guo W-z, Xue X-z, Wang L-c, Qiao X-j. Growth and quality responses of 'Green Oak Leaf' lettuce as affected by monochromic or mixed radiation provided by fluorescent lamp (FL) and light-emitting diode (LED). *Sciencia Horti* 2014;172:168-75.
- Cope K, Snowden MC, Bugbee B. Photobiological Interactions of Blue Light and Photosynthetic Photon Flux: Effects of Monochromatic and Broad-Spectrum Light Sources. *Photochem Photobiol* 2014;90:574-84.
- Cope KR, Bugbee B. Spectral effects of three types of white light-emitting diodes on plant growth and development: absolute versus relative amounts of blue light. *HortSci* 2013;48:504-09.
- Crocker W. Growth of plants. Twenty years' research at Boyce Thompson Institute. Growth of plants Twenty years' research at Boyce Thompson Institute 1948.

- Dougher TAO, Bugbee B. Differences in the response of wheat, soybean, and lettuce to reduced blue radiation. *Photochem Photobiol* 2001;73:199-207.
- Dougher TAO, Bugbee B. Long-term blue light effects on the histology of lettuce and soybean leaves and stems. *J Amer Soc Hort Sci* 2004;129:467-72.
- Folta KM, Childers KS. Light as a growth regulator: controlling plant biology with narrow-bandwidth solid-state lighting systems. *HortSci* 2008;43:1957-64.
- Folta KM, Maruhnich SA. Green Light: a signal to slow down or stop. *J Exp Bot* 2007;58:3099-111.
- Genty B, Briantais J-M, Baker NR. The relationship between the quantum yield of photosynthetic electron transport and quenching of chlorophyll fluorescence. *Biochimica et Biophysica Acta (BBA) - General Subjects* 1989;990:87-92.
- Goins G, Yorio N, Sanwo M, Brown C. Photomorphogenesis, photosynthesis, and seed yield of wheat plants grown under red light-emitting diodes (LEDs) with and without supplemental blue lighting. *J Exp Bot* 1997;48:1407-13.
- Goins GD, Ruffe LM, Cranston NA, Yorio NC, Wheeler RM, Sager JC. Salad crop production under different wavelengths of red light-emitting diodes (LEDs). SAE Tech Paper 2001.
- Hernández R, Kubota C. Physiological responses of cucumber seedlings under different blue and red photon flux ratios using LEDs. *Environ Exp Bot* 2015;DOI: 10.1016/j.envexpbot.2015.04.001.
- Hoenecke ME, Bula RJ, Tibbitts TW. Importance of 'blue' photon levels for lettuce seedlings grown under red-light-emitting diodes. *HortSci* 1992;27:427.
- Hogewoning SW, Douwstra P, Trouwborst G, Van Ieperen W, Harbinson J. An artificial solar spectrum substantially alters plant development compared with usual climate room irradiance spectra. *J Exp Bot* 2010b;61:1267-76.
- Hogewoning SW, Trouwborst G, Maljaars H, Poorter H, van Ieperen W, Harbinson J. Blue light dose-responses of leaf photosynthesis, morphology, and chemical composition of *Cucumis sativus* grown under different combinations of red and blue light. *J Exp Bot* 2010a;61:3107-17.
- Hogewoning SW, Wietjes E, Douwstra P, Trouwborst G, Van Ieperen W, Croce R, et al. Photosynthetic quantum yield dynamics: From photosystems to leaves. *Plant Cell* 2012;24.

- Hoover WH. The dependence of carbon dioxide assimilation in a higher plant on wave length of radiation. *Smithsonian Misc Collections* 1937;95:1-13.
- Hunt R. *Plant growth analysis*: Institute of Terrestrial Ecology; 1982.
- Inada K. Action spectra for photosynthesis in higher plants. *Plant Cell Physiol* 1976;17:355-65.
- Johkan M, Shoji K, Goto F, Hahida S, Yoshihara T. Effect of green light wavelength and intensity on photomorphogenesis and photosynthesis in *Lactuca sativa*. *Environ Exp Bot* 2012;75:128-33.
- Johkan M, Shoji K, Goto F, Hashida S-n, Yoshihara T. Blue light-emitting diode light irradiation of seedlings improves seedling quality and growth after transplanting in red leaf lettuce. *HortSci* 2010;45:1809-14.
- Kim H-H, Goins GD, Wheeler RM, Sager JC. Stomatal conductance of lettuce grown under or exposed to different light qualities. *Annals of Bot* 2004a;94:691-97.
- Kim HH, Goins G, Wheeler R, Sager JC, Yorio NC. Green-light supplementation for enhanced lettuce growth under red and blue LEDs. *HortSci* 2004b;39:1616-22.
- Kim HH, Wheeler R, Sager JC, Yorio NC, Goins G. Light-emitting diodes as an illumination sources for plants: A review of research at Kennedy Space Center. *Habitation* 2005;10:71-78.
- Klassen S, Ritchie G, Frantz J, Pinnock D, Bugbee B. Real-time imaging of ground cover: Relationships with radiation capture, canopy photosynthesis, and daily growth rate. *Digital imaging and spectral techniques: applications to precision agriculture and crop physiology* 2003:3-14.
- Lefsrud MG, Kopsell DA, Sams CE. Irradiance from distinct wavelength light-emitting diodes affect secondary metabolites in kale. *HortSci* 2008;43:2243-44.
- Leopold AC, Kriedemann PE. *Plant growth and development*. 2nd ed: McGraw-Hill, Inc.; 1975.
- Lin KH, Huang MY, Hauang WD, Hsu MH, Yang ZW, Yang CM. The effects of red, blue, and white light-emitting diodes on the growth, dvelopment, and edible quality of hydroponicly grown lettuce (*Lactuca sativa* L. var. capitata). *Sciencia Hortic* 2013;150.

- Massa GD, Kim HH, Wheeler RM, Mitchell CA. Plant productivity in response to LED lighting. *HortSci* 2008;43:1951-56.
- McCree KJ. The action spectrum absorptance and quantum yeild of photosynthesis in criop plants. *Agric Meteorol* 1972a;9:191-216.
- McCree KJ. Test of current definitions of photosynthetically active radiation against leaf photosynthesis data. *Agric Meteorol* 1972b;10:443-53.
- Morrow RC. LED lighting in Horticulture. *HortSci* 2008;43:1947-50.
- Nishio JN. Why are higher plants green? Evolution of the higher plant photosynthetic pigment complement. *Plant Cell Environ* 2000;23:539-48.
- Ouzounis T, Frette X, Rosenqvist E, Ottosen C. Spectral effects of supplementary lighting on the secondary metabolites in roses, chrysanthemums, and capanulas. *J Plant Physiol* 2014;171:1491-99.
- Ouzounis T, Parjikolaei BR, Fretté X, Rosenqvist E, Ottosen C-O. Predawn and high intensity application of supplemental blue light decreases the quantum yield of PSII and enhances the amount of phenolic acids, flavonoids, and pigments in *Lactuca sativa*. *Front Plant Sci* 2015;6.
- Paradiso R, Meinen E, Snel JF, De visser P, Van Ieperen W, Hogewoning SW, et al. Spectral dependence of photosynthesis and light absorpance in single leaves and canopy in rose. *Scientia Hortic* 2011;127:548-54.
- Poorter H, Remkes C. Leaf area ratio and net assimilation rate of 24 wild species differing in relative growth rate. *Oecologia* 1990;83:553-59.
- Quail PH. Phototsensory perception and signalling in plant cells: New paradigms? *Curr Opin Cell Biol* 2002;14:180-88.
- Sager JC, Smith WO, Edwards JL, L. CK. Photosynthetic efficiency and phytochrome photoequilibria determination using spectral data. *Trans ASAE* 1988;31:1882-89.
- Schuerger AC, Brown CS, Stryjewski EC. Anatomical features of pepper plants (*Capsicum annum* L.) grown under red light-emitting diodes supplemented with blue of far-red light. *Annals of Bot* 1997;79.
- Smith H, Whitelam GC. The shade avoidance syndrome: multiple responses mediated by multiple phytochromes. *Plant Cell Environ* 1997;20:840-44.
- Smith K. *The science of photobiology*: Springer Science & Business Media; 2013.

- Son K-H, Oh M-M. Leaf shape, growth, and antioxidant phenolic compounds of two lettuce cultivars grown under various combinations of blue and red light-emitting diodes. *HortSci* 2013;48:988-95.
- Stutte GW. Light-emitting Diodes for manipulating the phytochrome apparatus. *HortSci* 2009;44:231-34.
- Sullivan JA, Deng XW. From seed to seed: The role of photoreceptors in *Arabidopsis* development. *Dev Biol* 2003;260:289-97.
- Sun J, Nishio JN, Vogelmann TC. Green light drives CO₂ fixation deep within leaves. *Plant Cell Physiol* 1998;39:1020-26.
- Terashima I, Fujita T, Inoue T, Chow WS, Oguchi R. Green light drives leaf photosynthesis more efficiently than red light in strong white light: revisiting the enigmatic question of why leaves are green. *Plant Cell Physiol* 2009;50:684-89.
- Terfa MT, Solhaug KA, Gislerød HR, Olsen JE, Torre S. A high proportion of blue light increases the photosynthesis capacity and leaf formation rate of *Rosa* × *hybrida* but does not affect time to flower opening. *Physiol Plant* 2013;148:146-59.
- von Sachs J. Wirkungen farbigen Lichts auf Pflanzen. Halle; 1864.
- Wang XY, Xu XM, Cui J. The importance of blue light for leaf area expansion, development of photosynthetic apparatus, and chloroplast ultrastructure of *Cucumis sativus* grown under weak light. *Photosynthetica* 2014:1-10.
- Wang Y, Folta KM. Contribution of green light to plant growth and development. *Am J Bot* 2013; 100:70-78.
- Wang Y, Noguchi K, Terashima I. Photosynthesis-dependent and independent responses of stomata to blue, red, and green monochromatic light: Differences between the normally oriented and inverted leaves of sunflower. *Plant Cell Physiol* 2011;52:479-89.
- Wassink EC, Stolwijk AJ. Effects of light quality on plant growth. In: Blinks LR, editor. *Annu Rev Plant Physiol* 1956. p. 373-400.
- Wellburn AR. The spectral determination of chlorophylls a and b, as well as total carotenoids, using various solvents with spectrophotometers of different resolution. *J Plant Physiol* 1994;144:307-13.

Wheeler R, Mackowiak C, Sager J. Soybean stem growth under high-pressure sodium with supplemental blue lighting. *Agron J* 1991;83:903-06.

Yorio NC, Goins G, Kagie H, Wheeler R, Sager JC. Improving spinach, radish, and lettuce growth under red LEDs with blue light supplementation. *HortSci* 2001;36:380-83.









Yorio NC, Wheeler RM, Goins GD, Sanwo-Lewandowski MM, Mackowiak CL, Brown CS, et al. Blue light requirements for crop plants used in bioregenerative life support systems. *Life Support Biosph Sci* 1997;5:119-28.

APPENDICES

Appendix. Light levels

Table A1

Spectral characteristics of the eight treatments. Phytochrome photoequilibrium (PPE) and yield photon flux (YPF) as described by Sager et al. (1988).

| Light quality | SPECTRAL TREATMENT | | | | | | | |
|----------------|---|---|---|---|--|---|---|---|
| |  |  |  |  |  |  |  |  |
| Parameter | Red | Green | Warm ^{*†} | RB [†] | RGB ^{*†} | Neutral [*] | Cool [*] | Blue |
| PPF | 500 | 500 | 500 | 500 | 500 | 500 | 500 | 500 |
| YPF | 486 | 390 | 455 | 469 | 447 | 441 | 429 | 351 |
| YPF/PPF | 0.97 | 0.78 | 0.91 | 0.94 | 0.89 | 0.88 | 0.86 | 0.70 |
| PPE | 0.89 | 0.83 | 0.84 | 0.89 | 0.89 | 0.84 | 0.83 | 0.58 |

^{*}To minimize confounding spectral effects, only these four RGB, cool, warm and neutral white treatments were used in the blue light analysis.

[†]To minimize confounding spectral effects, only these three RGB, cool, warm and neutral white treatments were used in the green light analysis.