COMPARING THE EFFECTS OF GREENHOUSE SUPPLEMENTAL LIGHT SPECTRA ON THE

DEVELOPMENT AND YIELD OF HIGH-CBD HEMP

Project Report

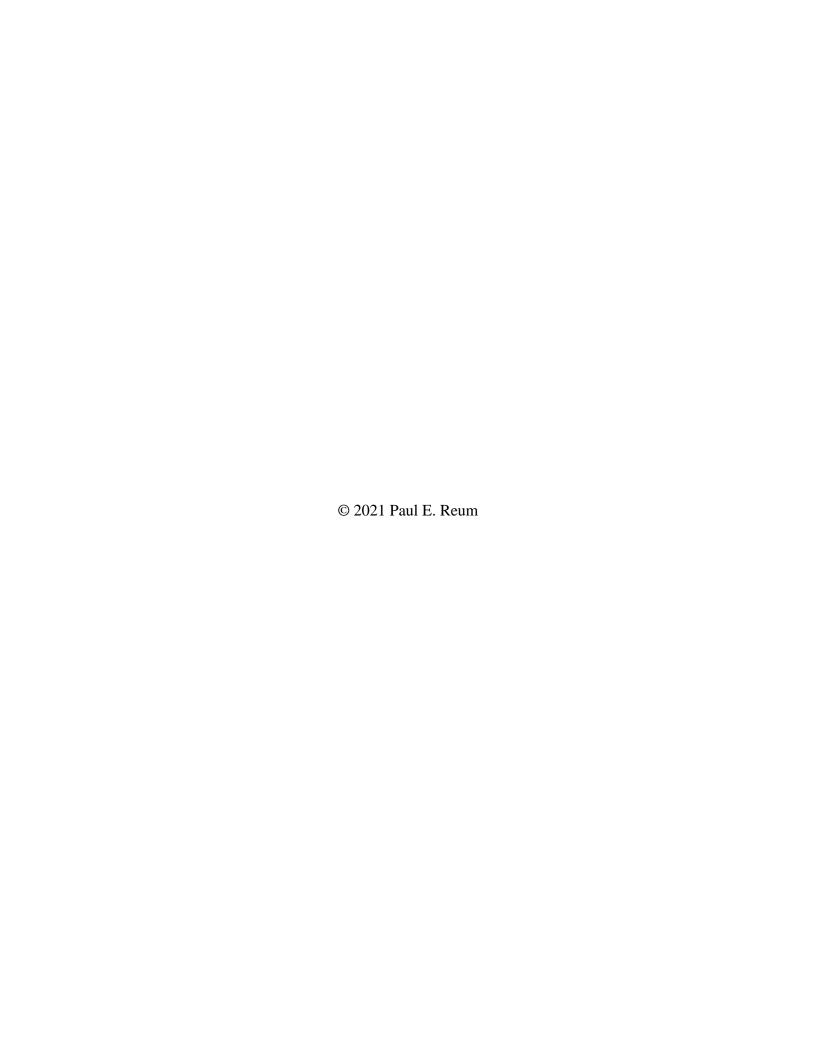
Presented to the Faculty of the Graduate School of Cornell University
in Partial Fulfillment of the Requirements for the Degree of
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by

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ABSTRACT

Over the past two decades, indoor production of *Cannabis sativa* (cannabis) has been rapidly expanding (Summers et al., 2021) (Wartenberg et al., 2021). Indoor production allows growers to control all aspects of the growing environment including cultivar selection, growing media, water use, nutrients, temperature, humidity control, lighting intensity and lighting quality. The optimized production results in a clean, high-quality final product that is suitable for the pharmaceutical industry or high-end market. Currently, indoor cannabis production is a highly energy intensive and ecologically unsustainable practice. Making a transition from sole-source indoor cannabis production (i.e. warehouse) to greenhouse cannabis production with supplemental lighting will result in lower production costs and lower greenhouse gas (GHG) emissions. Currently, there is an overall lack peer-reviewed data to support which supplemental lighting technologies lead to the greatest overall yields in greenhouse grown cannabis. Understanding which spectra of lighting are most beneficial to the development of cannabis in greenhouse production would inform producers on which types of lighting technologies they should invest in. Therefore, to address this gap in understanding, we performed a trial of six supplemental lighting treatments: 60:40 Red:Blue LED, 90:10 Red:Blue LED, Red:Blue LED with a Far-Red peak, high pressure sodium (HPS), Red:Blue LED with a UVA peak, and White LED. Two cultivars ('TJ's CBD' and 'T2'), were grown during their ten week flowering stage, and effects of light treatments were measured in terms of whole-plant wet and dry-weight, stem and leaf weight, flower weight, and final total cannabidiol (CBD) and tetrahydrocannabinol (THC) concentrations. Overall, the greatest lighting treatment effects were observed within the TJ's CBD cultivar, with fewer significant differences found within the T2 cultivar. With the exception of growth and height, the White LED treatment outperformed all other treatments. The White LED treatment led to the heaviest plants as well as increased flower yields and higher cannabinoid concentrations.

BIOGRAPHICAL SKETCH

Paul Reum, hails from Redford Michigan, a small township just outside of Detroit, Michigan. In his youth, Paul would spend most of his time outdoors exploring the neighborhoods around his in search of the small oases of nature that dotted Detroit's urban landscape. It was during these formative years that Paul found his love of nature and a passion that would later lead to him pursuing his Environmental and Sustainability Science degree from Cornell University in Ithaca, New York.

During his time at Cornell University, Paul has worked in both large-scale outdoor high-CBD hemp production, and greenhouse-controlled environment agriculture (CEA) production of high-CBD cannabis. Most recently, Paul has worked as a research technician for the Mattson Lab on several greenhouse crop experiments including lettuce, melons, tomatoes, strawberries, and hemp. In the past he has worked as a research technician studying the efficacy of biological control as a long-term solution for the invasive hemlock woolly adelgid as well as studying the dynamics of physical, hydrological, and biological properties of soil.

Paul was a Cornell Tradition fellow and served as a student advisory council member of the campus community service group. During his time as a tradition fellow, he personally led two student service trips to San Juan de la Concepcion, Nicaragua, where he and other tradition fellows worked on an organic farm, reforestation project and for a special needs school.

After speaking with the head of the Hemp Science Program at Cornell University, Dr. Carlyn Buckler about his interests in sustainability, energy-efficiency and cost-savings, Paul was paired with Dr. Neil Mattson's Lab at Cornell University in partnership with *GLASE*, the Greenhouse Lighting and Systems Engineering consortium.

ACKNOWLEDGEMENTS

First, I would like to thank my project advisor, Dr. Neil S. Mattson for his insight and guidance during this project. Neil provided me with a technician position in his lab, which helped me learn and gain practical experience. The Mattson Lab provided me with the plant material, greenhouse space and supplies I needed to complete this project. Neil was friendly, highly approachable, and made himself available anytime I needed assistance. Neil, along with the members of his lab made me feel accepted and like I belong in plant science at Cornell.

Second, Dr. Carlyn S. Buckler for seeing the potential in me and helping me identify my interest in controlled environmental agriculture (CEA) and cultivation. By recognizing my interests in CEA, Carlyn knew that connecting me with Neil Mattson and his lab team would lead to my success in the Hemp Sciences MPS program. Carlyn also worked to help find me job opportunities that fit my skill set and interests. Carlyn was always positive and believed in me even when I had doubts in myself.

Third, I would like to thank Nickolas Kaczmar. Nick provided me with the general framework needed to get the project started, as well as the expertise and advice to ensure the project's completion. Nick served as my direct supervisor in my technician position with the Mattson Lab. Under Nick's supervision, I have learned a great deal about greenhouse CEA production and gained invaluable hands-on experience that will prove highly beneficial in my chosen career path. Nick's knowledge, hard work, planning, support, and leadership skills were vital to the success of this project.

Fourth, I would like to thank Dr. Heather Grab who was a great help in getting the writing portion of this project completed. Heather helped me make sense of my statistical results and helped me present my data properly. Heather provided critical feedback as well as writing tools

and advice that allowed me to come up with a proper outline for the project. Heather helped me believe in my writing ability and reawakened my interest in scientific writing. Having regular meetings with Heather was essential to me finishing my writing goals during this project.

Fifth, I would like to thank my wife, Adrianne Chissus who was always there to support me and provide positive encouragement. Adrianne was always sympathetic and understanding of the amount of time I had to commit to my research and writing. She held it together when things got rough, and I will forever be grateful for her devotion and care.

Sixth, I would like to thank the other members of Neil Mattson's lab and the Greenhouse staff at Cornell's Guterman Bioclimatic Laboratories, especially Bret Timmons, who regularly oversaw the general care of my plants. Bret noticed my plants were getting top-heavy and took it upon himself to setup a trellis system that kept my plants upright through the remainder of the project. He also diagnosed a possible nutrient-burn issue before it became problematic, which was fundamental to the project having a positive outcome.

Last, I would like to thank Gavita Horticultural Lighting, Hawthorne Gardening Co., LumiGrow, and U.S. Global Resources Inc. This project required two light fixtures for each treatment, all of which were generously provided.

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LIST OF ABBREVIATIONS

Red to blue ratio of 60 percent red, 40 percent blue

90:10 Red to blue ratio of 90 percent red, 10 percent blue

CBD Cannabidiol

HID High Intensity Discharge

HPLC High Performance Liquid Chromatography

HPS High Pressure Sodium

HVAC Heating, Ventilation and Air Conditioning

LED Light Emitting Diode

PFD Photon Flux Density

PPFD Photosynthetic Photon Flux Density

THC Tetrahydrocannabinol

UVA Ultraviolet radiation in the 315-400 nm range

INTRODUCTION

Cannabis sativa L. (cannabis) is a versatile plant that provides cultivators with a variety of agricultural products such as grain, oils, fiber, cannabidiol (CBD), and tetrahydrocannabinol (THC). These products are used in a variety of industries, including the food, clothing, and pharmaceutical industries. With a recent rise in the demand for these cannabis products, cultivators are looking to find the most energy efficient and cost-effective options for growing high quality cannabis. Indoor (i.e. warehouse with lack of sunlight) cultivators have the benefit of increased pest protection and controlled environmental agriculture (CEA) in which nutrients, water use, transpiration and lighting can be optimized (van Iersel & Gianino, 2017). Additionally, drug type cannabis that is grown indoors sells for higher prices than cannabis grown outdoors due the consistency, cleanliness, and high yield of the final product (Chandra et al., 2017). However, indoor cannabis cultivation can be costly due to the increased capital costs as well as heating, ventilation, and air conditioning (HVAC) and lighting costs. Indoor horticultural operations are heavily dependent on electrical lighting, averaging between 5,200-6,500 operating hours per year for sole-source indoor production while supplemental lighting for greenhouse production averages 2,000 operating hours (Lee et al., 2020). Lighting is responsible for roughly 80% of the total electricity used (Magagnini et al., 2018) and is the second leading cause of total greenhouse gas (GHG) emissions (Mills, 2012, 2020; Summers et al., 2021) in the cannabis industry behind combined heating, cooling and humidity management (HVAC) needs (Summers et al., 2021). When focusing on the sustainability and economic viability of the cannabis industry moving forward, cultivators are looking for ways to lower their energy consumption and costs. Increasing the efficiency of grow lights and transitioning from traditional high-pressure sodium (HPS) lighting to light emitting diodes (LEDs) will lower operating costs

for the greenhouse CEA cannabis industry, especially for large-scale cultivators (Lee et al., 2020) (Singh et al., 2015). Many regions engaged in greenhouse cannabis cultivation will be reliant on supplemental lighting to maximize yields (Summers et al., 2021). Greenhouse cannabis production with optimized supplemental lighting could prove an invaluable tool that would allow cultivators to increase yields and long-term profits (Chandra et al., 2017)(Eaves et al., 2020a; Potter & Duncombe, 2012) while simultaneously reducing their GHG emissions (Mills, 2012)(Lee et al., 2020)(Summers et al., 2021).

Many crops show improved development and increased yields if provided the specific optimal spectrums of light (Both, 2000; Dueck et al., 2017; Legendre & van Iersel, 2021; Llewellyn et al., 2019; Särkkä et al., 2017; H. L. Smith et al., 2017). Early-stage cannabis studies (Backer et al., 2019; Danziger & Bernstein, 2021; Hawley et al., 2018a; Jenkins et al., 2021; Lalge et al., 2017; Magagnini et al., 2018; Mitchell Westmoreland et al., 2021; Rodriguez-Morrison et al., 2021; D. L. Smith et al., 2021) have yet to pinpoint the ideal spectrum for maximum flower yields and cannabinoid concentrations and have revealed conflicting results that need to be better elucidated.

Light quality, also sometimes referred to as spectral composition, is the relative number of photons of the blue, green, red, far-red and some ultraviolet portions of the spectrum provided to a given plant. Light quality is grouped into colors based on wavelength: 320-400 nanometers (nm) is UVA, 400-500 nm is blue, 500-600 nm is green, 600-700 nm is red, and 700-750 nm is far-red. Each of these wavelengths interact with special photoreceptors in the plants and cause a variety of physical and chemical responses within plants (Magagnini et al., 2018). Different light sources have varying degrees of light quality or spectra of light that they emit (Appendix 1a-5e).

HPS lighting is one of the most common and widely used types of lighting in the horticultural industry today. HPS lights are a type of High Intensity Discharge (HID) lamps that consume large amounts of electricity. The bulbs, reflectors, and ballasts of high-pressure sodium (HPS) lighting fixtures reach relatively high temperatures, which is a problem for greenhouse producers because higher bulb temperatures correlate to increased HVAC costs (Mills, 2012) (Summers et al., 2021). HPS fixtures also produce a lot of radiant heat directed toward the plant which may or may not be beneficial, depending on growing season. While HPS lamps weren't originally created for use in horticulture and indoor cropping systems, 96% of the overall light emitted by HPS lamps falls within the 400-700 nm photosynthetically active range making HPS lamps suitable for use as sole-source or supplemental lighting in horticultural applications (Magagnini et al., 2018). The greatest amount of the light emitted by an HPS falls within the 500-600 nm (green) range (Magagnini et al., 2018).

Currently, capital costs involved with LEDs compared to traditional HPS fixtures makes them a prohibitive choice for large scale greenhouse cultivation. However, newer LED technologies can offer a 35% or more reduction in energy consumption compared to traditional HPS lighting (Lee et al., 2020). LEDs offer a wide variety of light quality recipes that can be fine-tuned to the needs of a specific plant type or grow operation (Singh et al., 2015). White light is made up of a combination of different wavelengths that are present within the visible spectrum. Past studies (Mitchell Westmoreland et al., 2021) have shown that raising the blue fraction of light led to a 4.6% decrease in yield compared to HPS fixtures based on a per unit area basis, but the yield was 27% higher when applied on a per dollar of electricity cost. The supplemental addition of both blue and far-red LED light has been shown to stimulate growth and stem elongation in microgreens (Ying et al., 2020). Increased stem elongation could be a

sign that the plant is dedicating more resources into the development of stem material rather than bud or flower biomass. Stem elongation could play an important role in cannabis production as cultivators typically select for cultivars that maximize biomass allocation to inflorescence leading to increased yields. Another type of light that effects plant growth and physiology is light that falls within the UV spectrum. UV light is of special interest since a recent study (Rodriguez-Morrison et al., 2021) has shown that as UVB exposure increases, the weight and cannabinoid content of the inflorescence decreases, while conflicting previous research found a beneficial effect of UVB on cannabinoid concentration (Pate, 1983).

Light quantity or intensity can also impact the growth and development of cannabis, with higher light intensities showing a positive correlation among nearly all growth parameters of the plant, most importantly inflorescence yield, CBD, and THC concentrations (Eaves et al., 2020a; Hawley et al., 2018b; D. L. Smith et al., 2021). For this study, light intensity will be reported in photosynthetic photon flux density (PPFD) and can be defined as the amount of photosynthetically active radiation (PAR) that reached the canopy of our crop with units of μ mol·m⁻²·s⁻¹.

The overall objective of this study was to determine the influence of supplemental greenhouse lighting using high pressure sodium (HPS) and five unique LED treatments (90:10 R:B LED, 60:40 R:B LED, White LED, and R:B with Far-red and UVA peaks) on plant height, plant growth, fresh weight, dry weight, stem and leaf weight, flower weight, cannabidiol (CBD) percentage and tetrahydrocannabinol (THC) percentage of two different cultivars (T2 and TJ's CBD).

MATERIALS AND METHODS

Propagation, Establishment, and Growth

The two cultivars chosen for this study were "Trump 2" (T2) and "TJ's CBD" (TJ's CBD). T2 is a shorter, bushier plant with a wider architecture, and more fanned out leaves that produced numerous inflorescences of the same size (Appendix 2). TJ's CBD was taller with thin leaves and each plant had a distinct apical meristem (Appendix 2). For this study, the aim was to observe the same lighting treatment effects across two exceptionally diverse and genetically distinct cannabis varietals.

Cuttings of cultivars 'TJ's CBD' and 'T2' were procured from mother plants maintained by Dr. Neil Mattson's Laboratory at Guterman Bioclimatic Laboratories at Cornell University in Ithaca, NY. Shoot length for cuttings taken from the mother plants ranged between 10-16 cm in length. The cuttings were dipped in Clonex (Growth Technology Ltd., Taunton, Somerset, U.K., Hydrodynamics International Inc., Lansing, MI), placed into 3.8 x 3.8 cm rockwool cubes, and placed into 1020 trays. Trays were then placed into a misting system that provided a 10 second misting of water every 15 minutes. The cuttings received an 18-hour photoperiod that consisted of natural daylight as well as supplemental High-Pressure Sodium (HPS) lighting to avoid early flowering. Cuttings of both cultivars were maintained under these conditions in the propagation house Guterman GH 180 for 3 weeks.

Upon successfully rooting, the plants were transplanted into 10 cm pots with LM-111 all-purpose potting mix (LM-111, Lambert Peat Moss, Rivière-Ouelle, Canada). After transplanting, the plants were moved to grow-benches and provided with high pressure sodium (HPS) lights at 250 μmol·m⁻²·s⁻¹. All plants from both cultivars were provided a 18-hour photoperiod and watering took place on an as-needed basis with a 200 mg·L⁻¹ nitrogen fertilizer made with 15 N-

5 P - 15 K – 4 Ca – 2 Mg Jack's Professional LX Water-Soluble Fertilizer (JR Peter's Inc., Allentown, PA). Greenhouse temperatures were maintained with day/night temperatures of 25.5/16.5 °C. Due to less-than-ideal root development, a period of 21 days was needed before the plants could be transplanted into their final 11-liter (3 gallon) pots with LM-111. Plants were kept under the same temperature and light conditions as above for another 21 days to allow for adequate vegetative growth before the induction of flowering.

On February 17th, the plants were moved under their respective lighting treatments (described below), and flower induction was triggered by decreasing the photoperiod from 18 hours to 12 hours. Initial vegetative heights were taken of all plants. All other environmental factors remained constant throughout the experiment.

From April 7th to April 9th the plants were provided with a flush of clear water to address a possible nutrient burn issue. The flush was successful, and the experiment was able to move forward unimpeded.

Lighting Treatments

For this experiment, six total lighting treatments were applied for 70 days from February 17th until April 28th. The treatments used were as follows: HPS control (HS2000 600W, U.S. Global Resources, Florida, Texas), LED red to blue ratios of 90:10 and 60:40 (LumiGrow Pro 650e 585W, LumiGrow, Emeryville, California), a phosphor converted white LED (Gavita Pro 1700e 645W, Gavita, Vancouver, Washington) an experimental red:blue LED fixture which also included a Far-Red peak and an experimental red:blue LED fixture which also included a UVA peak. Each lighting treatment had two fixtures. Each light fixture was adjusted to produce as close to 200 µmol·m⁻²·s⁻¹ photosynthetic photon flux density (PPFD) as possible in the center of

the treatment area at one meter above the bench height. An Apogee PS-300 spectroradiometer (Apogee Instruments, Inc., Logan, Utah) recorded the spectrum and light intensity of each plant location in each treatment. Due to differences in lighting hardware some treatments received more or less than 200 µmol·m⁻²·s⁻¹ (Table 1). To ensure accuracy and that the only light source was from the light treatment fixture, initial measurements were taken at least 30 minutes after sunset. One-by-one, each lighting treatment was turned on and measurements for each plant location were taken. Each plant location was positioned to give the maximum amount of distance from its neighbors while remaining under their respective lighting treatment. All lighting treatments were given their own bench and two different cultivars (T2 and TJ's CDB) were placed under each treatment. Lighting treatments were applied to every other bench across one side of the greenhouse. Leaving an empty bench between treatments eliminated or minimized the chance of light from one treatment "bleeding" into a neighboring treatment. Three feet of black plastic was used to shade the White LED treatment and 4-inch-deep aluminum turkey baking trays were used to shade the control HPS from their respective neighboring treatments. Due to differences in genetics and overall growth patterns, all T2's were placed on the south-end of the benches closer to the exterior wall of the greenhouse, while TJ's CBD were placed toward the interior (north-end). This step was taken to avoid the potential for TJ's CBD to "shade-out" the shorter, bushier T2 plants. A layout of the experiment is provided in Figure 1. The average spectrum for each light treatment taken from the 7 plant locations is available in Appendix 1 and the average intensities are provided in Table 1. The white light in this experiment contained the greatest amounts of red, green, and blue light compared to other treatments (Appendix 1a-5e).

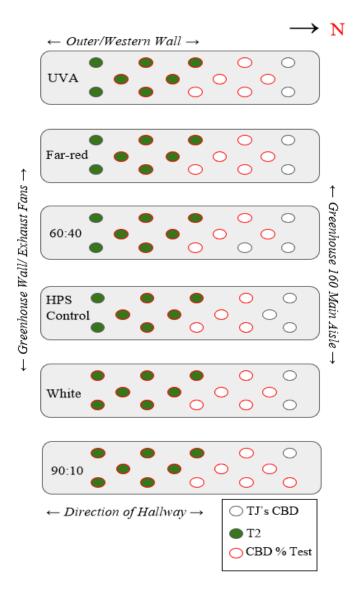


Figure 1: The layout of cultivars T2 and TJ's CDB under the experimental lighting treatments. Treatments were placed with an empty bench between them to reduce interference from neighboring treatments. Individual plants were spaced to allow maximum plant growth and minimal interference from neighboring plants but to ensure adequate lighting under the fixtures area of effect. Unfilled (white) circles represent TJ's CBD, and the dark-green filled circles represent T2. Plant numbers for each treatment were assigned as if reading from left to right, front to back (front being the main aisle of the greenhouse). The circles with red outlines indicate the plants that were chosen for HPLC analysis for each treatment. A two-foot buffer was included to allow for additional spacing away from the greenhouse exhaust fans.

Harvest Procedure

All plants were harvested on April 28th, which was 10 weeks (70 days) after the start of the short-day flower inducing photoperiods, in accordance with prior studies and industry standards (Oliver & McKeen, 2016). Prior to destructive harvesting, plants were chosen for cannabinoid analysis via high performance liquid chromatography (HPLC) based on the amount of light they had received over the course of the experiment. The plants which received the highest quantity of light from their respective lighting treatment were chosen for HPLC

cannabinoid analysis and are identified with a red circle in figure 1. None of the plants in the study were impacted by neighboring treatments. The top 10 centimeters of the apical meristem of the TJ's CBD hemp and the highest flower of the T2 hemp were removed, placed in a small, labelled paper bag and allowed to dry under the same conditions as mentioned above. Once the samples were dry, the plant material was placed into individual airtight containers for extractions and cannabinoid testing via High Performance Liquid Chromatography (HPLC). HPLC extractions were carried out in accordance with the procedures outlined in Toth et al., 2020 to determine the concentrations of Cannabidiol (CBD) and Tetrahydrocannabinol (THC).

Plant heights of all TJ's CBD plants were measured from soil-line to the apical meristem. Due to the different growth patterns of the T2 plants, their height was measured from the soil-line to the height of the highest fully developed flower (apical inflorescence). Plant branches were cut into 30-40cm segments, to allow for maximum airflow when placed into paper bags. All fresh plant material was weighed (fresh weight) on a scale to the nearest tenth of a gram.

Due to space limitations, we were unable to hang-dry the whole plants as is usually outlined in commercial applications. All samples were stored in open-top paper bags in two separate storage rooms that were outfitted with auxiliary ventilation capabilities due to Covid-19 requirements. The additional ventilation as well as added box fans allowed for the plants to dry in their bags at an ambient temperature ranging from 20-22°C and a relative humidity of 55-60%. The storage rooms were kept as dark as possible to avoid UV degradation to the cannabinoids. A selection of bags/plants were weighed on a scale every 48 hours until their weights remained constant, at which time they were considered dry. Plants were rotated in the bags every 48 hours to allow uniform drying across all of the plant material and to avoid moisture buildup and mold in the bottom of the bags. All plants were dry after 14 days of this drying method. Once dry, all

plants were hand trimmed and plant material was separated into stem & leaf material and flower material. The stem & leaf material and flower material were each weighed on a scale to the nearest tenth of a gram. Whole plant dry weights were calculated by summing the plant parts.

Experimental Design and Statistical Analysis

All statistical analyses were performed in RStudio version 4.0.3 (R Core Team, 2021, Vienna, Austria). An analysis of variance (ANOVA) was performed on heights, fresh weights, dry weights, total THC content and total CBD content with cultivar and treatment along with their two-way interaction as predictors. Post-hoc analyses among lighting treatment levels within each cultivar were conducted using Tukey's honestly significant difference test (alpha = 0.05). Results were plotted using RStudio.

RESULTS

The growth rate (gain in height from beginning to end of 10 week short-day photoperiod) of TJ's CBD was greater than the growth rate of T2 ($F_{(1,72)} = 714.2$, P < 0.0001). Lighting treatments had an effect on the growth rate of TJ's CBD but not T2 ($F_{(5,72)} = 5.8$, P =0.0001, Figure 2). The UVA lighting treatment had a greater growth rate than all other lighting types, while white light resulted in a lower growth rate than far-red (Figure 2). Plant height was significantly different only between cultivars ($F_{(1,72)} = 1921.4$, P < 0.0001) and there was no variation in response to lighting treatments within cultivars ($F_{(5,72)} = 1.1$, P = 0.359, Figure 3). Fresh weight of TJ's CBD was greater than the fresh weight of T2 ($F_{(1.72)} = 7749.9$, P < 0.0001). While trends in fresh weight among lighting treatments were similar ($F_{(5,72)} = 1.245$, P = 0.297), fresh weight only varied within the TJ's CBD cultivars (Figure 4). The mean dry stem and leaf weight was higher for all TJ's CBD than the mean dry stem and leaf weight of T2 ($F_{(1,72)}$ = 273.6, P < 0.0001). Dry stem and leaf weights in response to light treatment were similar ($F_{(5,72)}$ = 0.7, P = 0.620), and stem and leaf weights showed variation based on light source only within the TJ's CBD cultivar, with the White LED having a significantly greater stem and leaf weight than the 60:40 and 90:10 R:B LED treatments (Figure 5). Mean flower weight of TJ's CBD was greater than the mean flower weight of T2 for all lighting treatment levels ($F_{(1,72)} = 385.5$, P <0.0001). The lighting treatments had an impact on mean dry flower weight for TJ's CBD but not T2 ($F_{(5,72)} = 2.4$, P = 0.044). The HPS and White LED treatments had greater mean dry flower weights than the 60:40, 90:10, Far-red and UVA treatments (Figure 6). T2 had a greater total potential CBD (%w/w) than TJ's CBD ($F_{(1,72)} = 714.2$, P < 0.0001). Lighting treatments had an effect on both T2 and TJ's CBD ($F_{(5,72)} = 4.3$, P = 0.002, Figure 7). Within the T2 cultivar, White LED light had a higher total potential CBD concentration than the UVA lighting treatment

(Figure 7). Within TJ's CBD, White LED lighting had a greater total potential CBD concentration than the UVA, Far-red, 90:10 and 60:40 treatments (Figure 7) while the Far-red treatment showed a lower total CBD concentration that both the HPS control and White LED treatments (Figure 7). The total potential THC concentrations(% w/w) of the T2 cultivar were greater than the THC concentrations of TJ's CBD ($F_{(1,72)} = 74.2$, P < 0.0001). Lighting treatments showed an effect on TJ's CBD but not the T2 cultivar ($F_{(5,72)} = 3.7$, P = 0.005, Figure 8). Within the TJ's CBD cultivar, the White LED treatment had higher THC concentrations than 60:40, 90:10, Far-red and UVA treatments (Figure 8), while the Far-red treatment had lower THC concentrations than both the HPS control and White LED treatments (Figure 8).

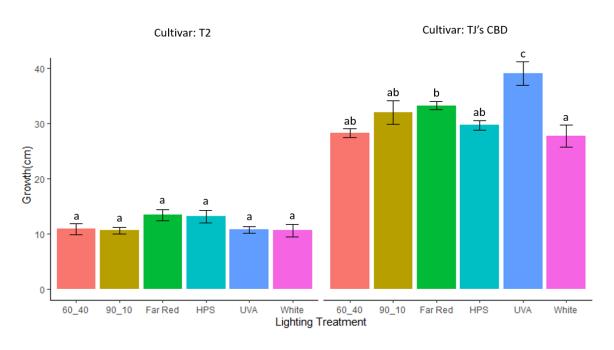


Figure 2: Mean specific plant growth in centimeters of 'T2' and 'TJ's CBD' hemp grown in a greenhouse under six lighting treatments for 70 days of short-day photoperiods prior to harvesting. Growth is defined as the difference between initial height measurements upon the start of short-day light periods and final heights measurements on the day of harvest. Data represent the means (± std. err.) of seven plants of each cultivar(14 total plants) per lighting treatment. Letters represent mean separation comparison using Tukey's HSD (alpha = 0.05). The lighting treatments consisted of two ratios of R:B LEDs (60:40 and 90:10), Far-Red LED, UVA LED, White LED and an HPS (high pressure sodium) control.

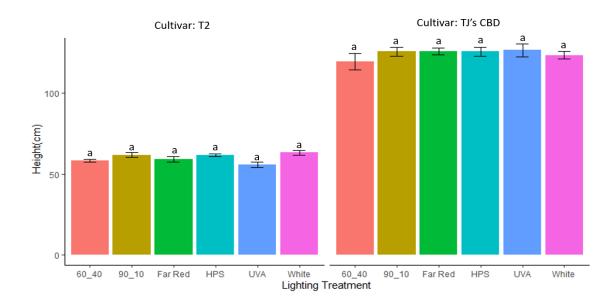


Figure 3: Mean final plant height in centimeters of 'T2' and 'TJ's CBD' hemp grown in a greenhouse under six lighting treatments for 70 days of short-day photoperiods prior to harvesting. Data represent the means (± std. err.) of seven plants of each cultivar (14 total plants) per lighting treatment. Letters represent mean separation comparison using Tukey's HSD (alpha = 0.05). The lighting treatments consisted of two ratios of R:B LEDs (60:40 and 90:10), Far-Red LED, UVA LED, White LED and an HPS (high pressure sodium) control.

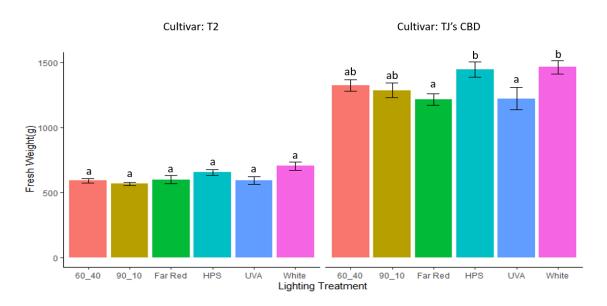


Figure 4: Mean whole-plant fresh weights of 'T2' and 'TJ's CBD' hemp grown in a greenhouse under six lighting treatments for 70 days of short-day photoperiods prior to harvesting. Data represent the means (± std. err.) of seven plants of each cultivar (14 total plants) per lighting treatment. Letters represent mean separation comparison using Tukey's HSD (alpha = 0.05). The lighting treatments consisted of two ratios of R:B LEDs (60:40 and 90:10), Far-Red LED, UVA LED, White LED and an HPS (high pressure sodium) control.

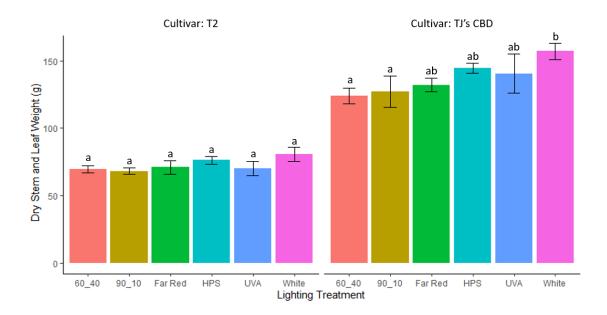


Figure 5: Mean dry stem and leaf weight in grams of 'T2' and 'TJ's CBD' hemp grown in a greenhouse under six lighting treatments for 70 days of short-day photoperiods prior to harvesting. Data represent the means (\pm std. err.) of seven plants of each cultivar (14 total plants) per lighting treatment. Letters represent mean separation comparison using Tukey's HSD (alpha = 0.05). The lighting treatments consisted of two ratios of R:B LEDs (60:40 and 90:10), Far-Red LED, UVA LED, White LED and an HPS (high pressure sodium) control.

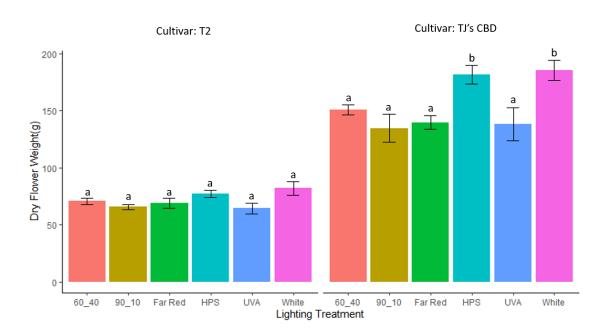


Figure 6: Mean Dry flower weight in grams of 'T2' and 'TJ's CBD' hemp grown in a greenhouse under six lighting treatments for 70 days of short-day photoperiods prior to harvesting. Data represent the means (± std. err.) of seven plants of each cultivar (14 total plants) per lighting treatment. Letters represent mean separation comparison using Tukey's HSD (alpha = 0.05). The lighting treatments consisted of two ratios of R:B LEDs (60:40 and 90:10), Far-Red LED, UVA LED, White LED and an HPS (high pressure sodium) control.

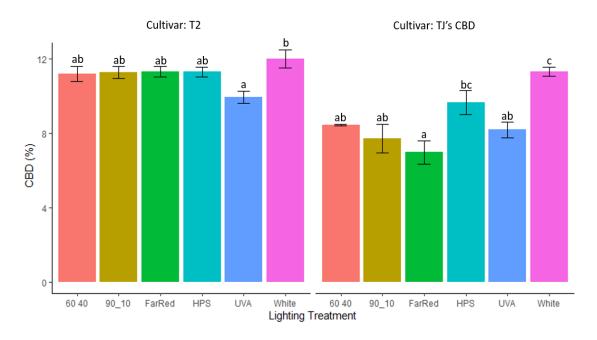


Figure 7: Mean total potential CBD concentrations (% w/w) of 'T2' and 'TJ's CBD' hemp grown in a greenhouse under six lighting treatments for 70 days of short-day photoperiods prior to harvesting. Data represent the means (\pm std. err.) of seven plants of each cultivar (14 total plants) per lighting treatment. Letters represent mean separation comparison using Tukey's HSD (alpha = 0.05). The lighting treatments consisted of two ratios of R:B LEDs (60:40 and 90:10), Far-Red LED, UVA LED, White LED and an HPS (high pressure sodium) control.

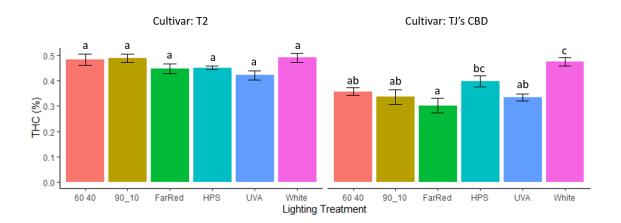


Figure 8: Mean total potential THC concentrations (% w/w) of 'T2' and 'TJ's CBD' hemp grown in a greenhouse under six lighting treatments for 70 days of short-day photoperiods prior to harvesting. Data represent the means (± std. err.) of seven plants of each cultivar (14 total plants) per lighting treatment. Letters represent mean separation comparison using Tukey's HSD (alpha = 0.05). The lighting treatments consisted of two ratios of R:B LEDs (60:40 and 90:10), Far-Red LED, UVA LED, White LED and an HPS (high pressure sodium) control.

DISCUSSION

Research on the effects of supplemental lighting on greenhouse cannabis is currently in its infancy. This study as well as follow-up studies will be instrumental in establishing supplemental lighting recommendations for greenhouse cannabis producers in the future. The data obtained from this study indicates that White LEDs and HPS lights are the most suitable options for supplemental lighting in greenhouse cannabis production. White LEDs and HPS lamps resulted in the greatest fresh weights, dry flower weights, CBD concentrations and THC concentrations. However, due to variability in light intensity delivered by treatment, the impact of light intensity rather than light spectrum cannot be discounted, and further experimentation is required.

Due to differences in cultivar genetics, the study found significant differences between cultivars on all measured variables. Apart from CBD content, T2 did not show any significant effects on measured parameters based on lighting quality. TJ's CBD showed much greater genetic plasticity in its response to light quality and exhibited quantifiable differences in all measurements except final plant height. This shows that cultivar selection is highly important when aiming to maximize yields.

For this study, we measured plant growth as the difference between initial plant height and the plant height on the day of harvest. In this measure, UVA outperformed all other lighting treatments. Secondarily, the far-red treatment performed better than the White LED treatment. This falls in line with past research (Legendre & van Iersel, 2021; Zhen & van Iersel, 2017) that shows far-red light is needed for efficient photosynthesis and can increase both the leaf area and canopy size of plants, which could in turn lead to an increased growth rate we see in our study.

The UVA treatment showed the greatest overall plant growth (height gain) compared to all other treatments, which is peculiar considering past research (Rodriguez-Morrison et al., 2021) showed increased UV levels had a negative influence on cannabis growth in terms of both biomass and height. The UVA lamps used in our study may have caused a "stretching" effect in the plants due to their lower overall low intensity (Mattson, 2021). TJ's CBD plants placed under the UVA treatment had a mean initial plant height of 87.6 cm, while the mean initial plant heights of the White LED and HPS treatments was 96.0 cm and 96.1 cm respectively, showing some disparity in the initial mean heights of plants placed under differing treatments. This study revealed significant cultivar-specific genetic effects on the final heights of cannabis with T2 average plant height of 59.9 cm and TJ's CBD average plant height of 124.2 cm. However, the study found no significant differences in the final heights of cannabis based on light quality (Figure 3), unlike previous studies (Danziger & Bernstein, 2021) that reported a 1:4 red to blue ratio LED resulted in taller plants when compared to HPS, 1:1 red to blue LED, and white LED treatments. Therefore, the UVA results in this study could be superficially attributed to the fact that the UVA treatment started with shorter average plants. Further testing with a more homogeneous (i.e., matching initial heights) crop could help to explicate this issue.

Whole plant fresh weight was greatest for the HPS and White LED treatments with UVA and far-red treatments having the lowest fresh weights. HPS and White LED had mean fresh weights of 1446.9 g and 1465.3 g, while far-red and UVA had mean fresh weights of 1216.9 g and 1222.9 g respectively. These results show that supplemental HPS and White LEDs have the potential to increase yields by more than 200 g per plant, compared to the far-red and UVA treatments, which could prove highly profitable to the cannabis industry. Unlike all other treatments, the HPS and White LEDs contain relatively large amounts of green light. Green light

plays an important role in carbon assimilation and biomass accumulation in the lower canopy of plants and provides positional signals that allow plants to better adapt to their lighting environment (H. L. Smith et al., 2017). It should be noted that HPS and White LED also delivered the greatest intensity of light, therefore more research should be done to separate the impact of light quality and light quantity.

In this study, dry stem and leaf weight (non-flower biomass) of TJ's CBD was greater for the White LED treatment compared to the 60:40 red to blue ratio LED and 90:10 red to blue ratio LED treatments. Previous studies (Lalge et al., 2017) (Magagnini et al., 2018) show plants that were provided white LED light showed increased height, leaf area and stem elongation compared to plants grown strictly under red and blue LEDs.

Dry flower weight was greatly increased for the HPS, and White LED treatments compared to all other treatments. This result is supported by Eaves et al., 2020 that showed an increase in inflorescence yield and flower density that corresponded with an increase in white LED light and is also supported by (Magagnini et al., 2018) that showed an increase in yield with HPS compared to all LED treatments.

In this study, HPLC extractions were employed to determine the concentrations (% w/w) of total potential CBD and THC present in the apical inflorescence of a subset of plants. HPLC results confirmed lighting effects on CBD concentrations across both cultivars. T2 showed White LED improved CBD concentrations compared to the UVA (Figure 7). TJ's CBD showed increased CBD concentrations under the White LED treatment compared to all other treatments, but also established that HPS has improved CBD concentrations, compared to far-red. The HPLC results confirmed lighting effects on THC concentrations for TJ's CBD only. The results for THC follow a very similar trend to CBD concentrations (Figure 7, Figure 8).

It is important to note that due to technological differences between our lighting fixtures, we were unable to get a matching photosynthetic photon flux density (PPFD) across all treatments of the experiment. The HPS and White LED treatments had the highest light intensities, averaging a greater photosynthetic photon flux density (PPFD) than all other lighting types (Table 1). Scientific literature has shown that cannabis yield increases linearly with PPFD, increasing to as high as 1,500 μmol·m⁻²·s⁻¹ (Eaves et al., 2020b) and that a higher PPFD environment increases the size, quality, and density of cannabis inflorescence (D. L. Smith et al., 2021). Normalizing PPFD across all treatments in future studies will help determine whether it was the quality of the HPS and White LED spectra or the light intensity that was causing the increase in biomass compared to all other treatments. A study by (Eaves et al., 2020a) demonstrated that high intensity LEDs such as our White LED treatment can produce up to 0.77 grams/watt of electricity when compared to traditional high intensity HPS lamps that produced 0.35 grams/watt, suggesting that high intensity, broad spectrum, White LED fixtures may be the most economic and efficient option for greenhouse cannabis production.

In the statistical analysis of this experiment numerous clear trends in the data could be visually identified but were not substantiated due to a smaller-than-ideal sample size. To increase the statistical significance of these trends and our other results, it is suggested that additional replicates with larger sample sizes be run in the future. Furthermore, running this experiment concurrently against a sole-source indoor lighting experiment could benefit future comparisons between greenhouse grown cannabis and growth chamber cannabis. Including subcanopy lighting in future experiments could help us with further optimization as this has been shown to improve inflorescence yield and alter cannabinoid profiles (Hawley et al., 2018b).

The overall objective of this study was to identify the impact of supplemental lighting spectra on the growth, development and yield of cannabis and determine which lighting type is most suitable for the greenhouse production of cannabis. Initial findings suggest that there is a difference in cannabis yield based on light quality and light intensity.

CONCLUSION

Currently, research on both the quality and quantity of lighting on drug type cannabis is lacking, especially as it pertains to supplemental greenhouse lighting. This experiment showed that supplemental light quality can influence the growth, fresh whole-plant weight, dry stem and leaf weight, dry flower weight, THC concentrations and CBD concentrations of high-CBD cannabis grown in a greenhouse. In this experiment, nearly all significant results were found within the TJ's CBD cultivar which showed a greater plasticity to environmental differences than the T2 cultivar. Overall growth of TJ's CBD was greatest for the UVA treatment compared to all other treatments. Whole-plant fresh weight of TJ's CBD was greater for the HPS, and White LED treatments compared to the Far-red and UVA. Dry stem and leaf weight (non-flower biomass) of TJ's CBD was greater for the White LED treatment compared to the 60:40 red to blue ratio LED and 90:10 red to blue ratio LED treatments. Dry flower weight of TJ's CBD was considerably greater for the HPS, and White LED treatments compared to all other treatments. Concentration of CBD (% w/w) of T2 was greater for the White LED over the UVA treatment while concentration of CBD of TJ's CBD was greater for the White LED treatment compared to all other treatments. Concentrations of THC in TJ's CBD were greatest for the White LED treatment compared to all other treatments. Future experiments would help to substantiate this study's results. Better monitoring of light intensity will ensure that results can be properly attributed to spectra and light quality effects rather than light quantity effects. Improved understanding of the effects of supplemental greenhouse lighting on cannabis production will allow for an easier transition for indoor cannabis producers who are looking to increase profits while reducing their adverse environmental impact.

APPENDIX

Table 1. The average light intensity $(\mu mol \cdot m^{-2} \cdot s^{-1})$ each treatment was receiving. Data represent the average of fourteen plant locations.

Lighting Treatment	Average intensity (μmol·m ⁻² ·s ⁻¹)	Standard deviation
90:10	145.7764	48.16509
White LED	223.4128	72.31973
HPS	241.3087	36.03606
60:40	136.2295	48.72198
Far-Red	112.9463	30.55876
UVA	125.956	53.24999

Appendix 1: Light Spectrum of Supplemental Lighting Treatments

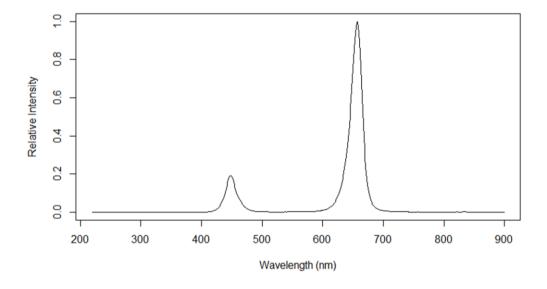


Figure 1a: Light Spectrum (relative intensity in photon flux density) from 220 to 900 nm for the 90:10 R:B lighting treatment. Data represent the average of fourteen plant locations.

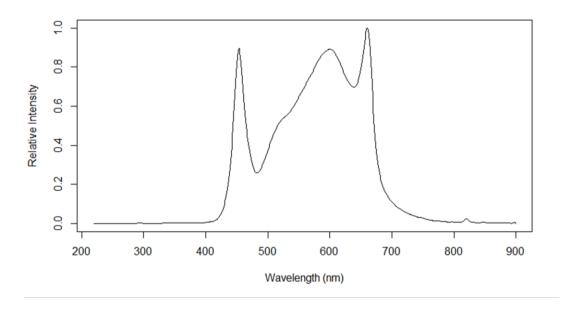


Figure 1b: Light Spectrum (relative intensity in photon flux density) from 220 to 900 nm for the White LED lighting treatment. Data represent the average of fourteen plant locations.

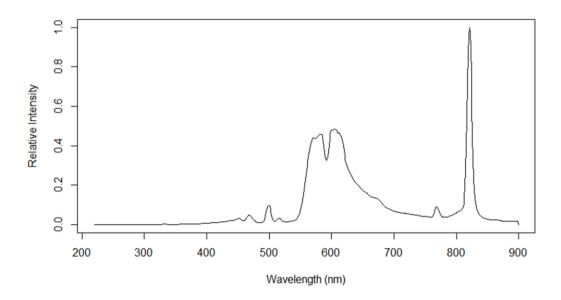


Figure 1c: Light Spectrum (relative intensity in photon flux density) from 220 to 900 nm for the HPS Control lighting treatment. Data represent the average of fourteen plant locations.

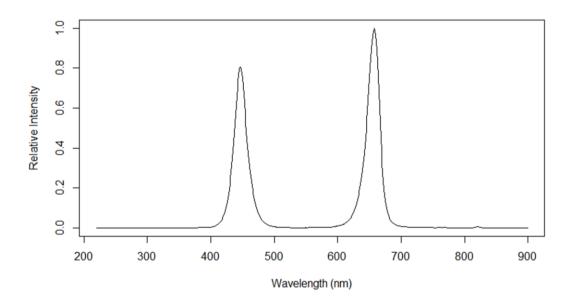


Figure 1d: Light Spectrum (relative intensity in photon flux density) from 220 to 900 nm for the 60:40 R:B lighting treatment. Data represent the average of fourteen plant locations.

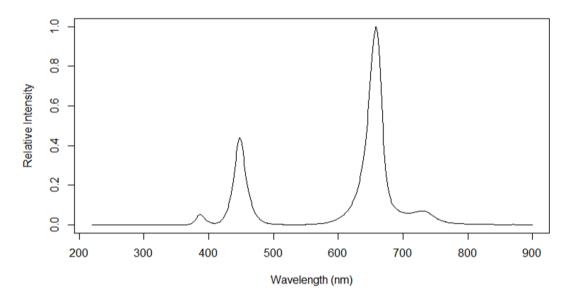


Figure 1e: Light Spectrum (relative intensity in photon flux density) from 220 to 900 nm for the Far-Red lighting treatment. Data represent the average of fourteen plant locations.

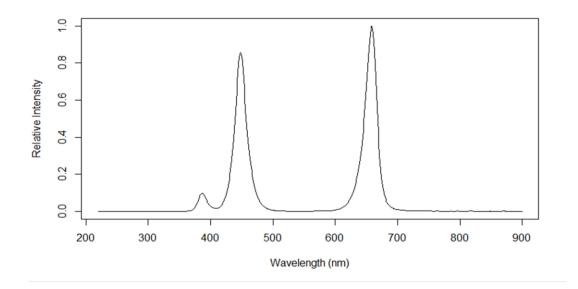


Figure 1d: Light Spectrum (relative intensity in photon flux density) from 220 to 900 nm for the UVA lighting treatment. Data represent the average of fourteen plant locations.

Appendix 2: Images of morphological differences between TJ's CBD and T2.



Figure 2a: Initial differences in morphological traits and initial plant heights TJ's CBD (left) and T2 (right).



Figure 2b: Mid-study differences in morphological traits and plant heights TJ's CBD (left) and T2 (right).



Figure 2c: End of study differences in morphological traits and initial plant heights TJ's CBD (left) and T2 (right).



Figure 2d: Plants under 90:10 R:B LED treatment TJ's CBD (left) and T2 (right).



Figure 2e: Plants under White LED LED treatment TJ's CBD (left) and T2 (right).



Figure 2f: Plants under high pressure sodium HPS treatment TJ's CBD (left) and T2 (right).



Figure 2g: Plants under 60:40 R:B LED treatment TJ's CBD (left) and T2 (right).



Figure 2h: Plants under Far-Red LED treatment TJ's CBD (left) and T2 (right).



Figure 2i: Plants under UVA LED treatment TJ's CBD (left) and T2 (right).

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Rubric for Evaluation of MPS Student Progress

Student Name: Paul Reum

Advisor Name: Neil Mattson

Date: 15 December 2021

The purpose of this evaluation is 2-fold: 1. To monitor the performance and progress of our students and 2. To develop evidence for assessing the quality of the MPS program overall. Advisors may use grades, conversations with the student, the project outline, the project report, and other observations to make their assessments. This evaluation should be turned in to the Graduate Field Office at the end of each semester for each student. It is up to the advisor whether or not to share the evaluation with the student.

Choose rating (1, 2, 3, or 4) that applies for each outcome category

Graduate Education Outcomes The student will be able to:	1 (Unacceptable)	2 (Fair)	3 (Very Good)	4 (Outstanding)
Demonstrate knowledge of appropriate subdiscipline(s) of food science.	Gaps in basic knowledge. Does not understand basic concepts, processes, or conventions of the discipline.	Displays a basic understanding of the field.	Displays a solid understanding of the field. Some exploration of interesting issues and connections.	Demonstrates thorough mastery as well as creativity in drawing on multiple sources. Synthetic and interdisciplinary. Demonstrates a deep understanding of the discipline.
Show effective oral communication skills.	Argument is weak, inconsistent, contradictory, unconvincing or invalid.	Provides solid, expected results and answers. Clear and coherent.	Gives a solid argument with novel or fresh insights. Original with clear and coherent details.	Compelling, exciting, and persuasive. Has a point of view and a confident, independent, authoritative voice.
Respond adequately to questions posed.	Unable to articulate an argument.	Provides a coherent response with some logic gaps or inconsistencies.	Shows understanding and mastery of subject matter.	Exhibits mature, independent thinking. Demonstrates command and authority over the material.
Display effective written communication skills.	Academic writing lacks structure and organization. Writing has extensive spelling and grammatical errors.	Writing is adequate. Structure and organization are weak but sufficient.	Well written and well organized.	Concise, elegant, engaging, interesting, sophisticated, and original. Connects components seamlessly.
Effectively frame or communicate the student's project.	No project. Question or problem is trivial, weak, unoriginal, or previously solved.	Demonstrates competence but is not very original or significant. Displays little creativity, imagination, or insight.	Has a compelling question or problem. Argument is strong, comprehensive, and coherent. Has some original ideas, insights, and observations.	Argument is focused, logical, rigorous, and sustained. Proposed project is original, ambitious, creative, significant, and thoughtful. Asks new questions or addresses an important question or problem.

Rubric and evaluation form for MPS Project Report

Student Name: Paul Reum

Advisor Name: Neil Mattson

Date: 15 December 2021

The purpose of this evaluation is to develop evidence for assessing the quality of the MPS project. This evaluation should be turned in to the Graduate Field Office after completion of the MPS Project Presentation for each student. It is up to the advisor whether or not to share the evaluation with the student.

Choose rating (high pass, pass, low pass, fail, no information) that applies for each outcome category

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	HP	Р	LP	F	n/i
The MPS project report is	Χ				
 Formatted in a manner appropriate to the 					
discipline					
 Uses citations correctly and effectively 					
 Is written in a professional style 					
Project objective and goals are well-defined and clearly	Χ				
stated.					
Literature review is current, comprehensive, and	Χ				
provides the relevant context for project report.					
Literature is synthesized and evaluated critically in a	Χ				
manner that demonstrates a comprehensive					
understanding of the issue and its significance.					
Tables and figures are used effectively.	Χ				
Project report applies a critical perspective to the issue		Χ			
and draws appropriate conclusions stating the strengths,					
weaknesses, and limitations of the report and the					
conclusions					
Conduct of project report and use of literature meets	Χ				
ethical standards.					