Light Spectra Have Minimal Effects on Rooting and Vegetative Growth Responses of Clonal Cannabis Cuttings

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Abstract. Until recently, most clonal cannabis (Cannabis sativa) has been propagated using fluorescent lights. Transitioning to light-emitting diodes (LEDs) may be a viable alternative to fluorescent lighting, enabling cultivators to provide specific spectrum treatments to enhance rooting while also saving energy. Vegetative stem cuttings of ‘Gelato-27’, ‘Grace’, and ‘Meridian’ were rooted for 15 days under various combinations of blue (B), red (R), ultraviolet-A (UVA) LEDs, phosphor-converted white (W) LEDs, and a fluorescent (F) control treatment, each with a canopy-level photosynthetic photon flux density (PPFD) of 200 μmol·m⁻²·s⁻¹ and 16-hour photoperiod. The photon flux ratios of blue (B; 400–500 nm) and red (R; 600–700 nm) narrowband LED treatment combinations were (1) BR, fixed spectrum of B15:R85; (2) B, B75:R25 on day 0–2 followed by B15:R85 on day 2–14; (3) B+UVA, B75:R25 on day 0–2 followed by B15:R85 on day 2–14 plus 15 μmol·m⁻²·s⁻¹ of UVA on day 7–14; (4) B50, B15:R85 on day 0–7 followed by B50:R50 on day 7–14. The W and F treatments both had static spectra. After the propagation period (i.e., plug stage), a portion of the cuttings under each treatment × cultivar combination were destructively harvested and the remainder were transplanted and grown vegetatively for an additional 21 days (i.e., transplant stage) until treatment × cultivar combination were destructively harvested and the remainder were.

Drug-type cannabis is an economically valuable crop that is often grown in controlled environments (CE), such as greenhouses with supplemental lighting or indoors with 100% electrical lighting, as these environments can produce higher quality crops than outdoor production. CE production allows cultivators to grow cannabis year-round with less concern about sub-optimal weather, including stabilizing crop light exposure and the avoidance of adverse weather (Zheng 2021). Further, CE production enables greater control of the day-length, which is a key production parameter for both inhibiting and inducing flowering in commercial cannabis production, depending on the growth stage. The use of electric lighting in indoor cannabis cultivation facilitates the production of uniform plants with optimal (for cultivators’ individual production protocols) morphological attributes (Moher et al. 2022; Potter 2014).

Cannabis production begins with a propagation period to establish transplants. Propagation can be accomplished via seed (Farag and Kayser 2017), tissue culture (Wang et al. 2009), or clonal cuttings from vegetative tissues (Campbell et al. 2019; Caplan et al. 2018). Most commercial CE cannabis cultivators opt for clonal stem cuttings since they can be harvested from stock mother plants and can provide uniform transplants with predictable growth and cannabinoid composition (Coffman and Gentner 1979; Potter 2009; Small 2017). The canopy-level PPFD [400–700 nm (μmol·m⁻²·s⁻¹)] is usually low during this stage (Fluence 2020; Lumigrow 2017) to minimize water stress as cuttings establish a root system. Once sufficiently rooted, plants are transplanted into larger containers to provide space for the root systems to enlarge appropriately to support the plant through the subsequent vegetative and flowering stages. Robust root systems help cuttings withstand transplant shock and endure the higher light intensities that are commonly used in the vegetative stage (Fluence 2020; Lumigrow 2017), especially under increasing transpiration loads associated with rapid vegetative growth.

The CE horticulture industry has adapted electric lighting technologies that were initially developed for human vision, such as high-pressure sodium, metal halide, and fluorescent for use in various crop production capacities. These technologies are each characterized as having fixed spectral output but with very disparate spectrum distributions (Dutta Gupta and Agarwal 2017). Importantly, the spectral output of these traditional technologies has not been optimized for light utilization by plants. Of these technologies, fluorescent lighting is used most commonly in indoor propagation systems (Bilodeau et al. 2019) because it is economical at a low intensity and is amenable to use in multitiered production systems that maximize space use efficiency. Recently, cultivators have been opting for LED technologies to replace the traditional horticultural lighting technologies because of their longer lifetimes, higher energy efficiency, and ability to provide customized spectra that are designed specifically for plant production (Nelson and Bugbee 2014; Singh et al. 2015; Yeh and Chung 2009). Customized spectra leverage the narrowband emissions of different diode colors by varying the proportions of different types of LEDs within a fixture. Further, some fixtures leverage the inherent dimming capabilities of LEDs to enable dynamic spectral control at a fixture level by manipulating the photon flux of individual LED color channels within a given fixture.

Combinations of light from blue (B, 400–500 nm) and red (R, 600–700 nm) wavebands commonly dominate the spectral output of horticultural LED fixtures. This is because B and R wavelengths have been reported to have the highest photosynthetic quantum yield (McCree 1972) and combinations of B and R do not often elicit abnormal morphological or physiological responses (Bantis et al. 2018; Kong et al. 2018). Further, photons from R wavelengths often comprise 75% to 95% of the photon flux in B+R spectra (Jones-Baumgardt et al. 2019; Kong and Zheng 2018; Randall and Lopez 2014; Ying et al. 2020), because R photons are more
energy efficient to produce than B (Kusuma et al. 2020). The magenta-hued environments resulting from B+R spectra may be uncomfortable to work under and can make it difficult to evaluate crop health. Therefore, phosphor-converted light from “white” LEDs are also often added to B+R to provide broader spectrum distributions, despite their inherently lower efficacy (Kusuma et al. 2020). By design, white LEDs already contain a significant amount of B, so narrowband B LEDs may not be used in these spectrum combinations.

Given the prescribed advantages of horticultural LED technologies, there is a need to evaluate the efficacy of using the various spectral combinations available with LEDs, in comparison with fluorescent lighting, for the propagation phase of clonal cannabis. Just as optimal light intensity levels can vary at different stages of production (Moher et al. 2022; Rodriguez-Morrison et al. 2021), the ideal light spectrum distribution may depend on a crop’s growth stage. For example, the use of high proportions of B at the start of clonal propagation may promote faster root initiation (Gil et al. 2020). The addition of UVA (315–400 nm) may increase root biomass and make the plants more resistant to transplant stress (Verdaguer et al. 2017). Adding higher proportions of B proximal to transplant may aid with promoting compact morphology (i.e., short internodes) of transplants (Ying et al. 2020). With the capability of enhanced LED technologies to modify the spectral output, it is possible that a lighting protocol that uses a temporally-dynamic spectrum (i.e., varying the photon flux ratios over time) may enhance the rooting performance of cannabis cuttings vs. a fixed spectrum, whether the fixed spectrum is provided by fluorescent or LED sources. Since there is limited scientific information related to lighting during the propagation of clonal cannabis, the effects of spectrum during this dynamic phase of plant development is unknown.

The overall objective of this study was to compare a range of LED spectrum treatments, both fixed and temporally dynamic, with fluorescent lighting for the propagation stage of clonal cannabis. The overall hypothesis is that some LED spectral combinations, either fixed or dynamic, will be superior to fluorescent lighting for producing cannabis transplants. Specific objectives were 1) compare fixed-spectrum distributions that are commonly used in horticultural LEDs (e.g., B mixed with R, phosphor-converted W) with fluorescent lighting for the rooting, growth, and transplant performance of clonal cannabis cuttings; and 2) investigate the effects of temporally-dynamic vs. fixed LED and fluorescent spectrum treatments on the rooting, growth, and transplant performance of clonal cannabis cuttings.

Materials and Methods

Plant material. Uniform clonal vegetative stem cuttings (hereafter referred to as cuttings) of three cannabis cultivars were used as treatment plants: ‘Gelato-27’ (short, bushy, and slow rooting), ‘Grace’ (relatively short and moderate rooting), and ‘Meridian’ (tall, stretched, and fast rooting). Cuttings of ‘Gelato-29’ were used as border plants in all plots. All cuttings were taken from 3-month-old mother plants that had been grown indoors under a PPFD of 400 μmol·m⁻²·s⁻¹ provided by metal halide fixtures (NXT-LP CMH; PL Light Systems Inc, Beamsville, ON, Canada), 18/6 h on/off photoperiod, 65% relative humidity (RH), and 24–20°C light/dark. Cuttings with three to four nodes and 10 to 16 cm in length were taken using sterilized scissors, and basal ends were cut on a 45-degree angle and scoured using a sterilized scalp. The basal end of each cutting was coated with rooting hormone (StimRoot #1; Master Plant Prod-Inc., Brampton, ON, Canada) and then inserted into a cylindrical rockwool plug (3.6-cm diameter × 4.0-cm height; Grodan, Milton, ON, Canada) contained in 50-cell propagation trays (0.5 × 0.3 m, 50 Plug Pre-filled; A.M.A Horticulture Inc., Kingsville, ON, Canada). The rockwool plugs were presoaked in a preventive biological fungicide (RootShield WP; Bioworks, Victor, NY, USA) at 0.45 g·L⁻¹ in sterile reverse osmosis (RO) water, resulting in an electrical conduc-

ity (EC) of 0.7 ds·m⁻¹ and a pH of 5.2. The propagation trays were covered with transparent vented plastic lids (7-inch Propagation Dome; Mondi Products, Vancouver, BC, Canada) and placed in flats without drainage holes (NR Open Flat; FloraPlast, Chatham, ON, Canada).

Growing conditions. The experiment was conducted in a commercial cannabis production facility in Southern Ontario, Canada. Three growth enclosures (5.9 × 4.1 × 2.7 m) were constructed, each consisting of two benches (5.9 × 1.8 m) that were separated by 0.5 m and encompassed with panda film (Vivosun, City of Industry, CA, USA), with the black side facing inward, to block ambient light from entering the enclosures (Fig. 1). Each enclosure was divided into six plots (three plots per bench), each equipped with its own light fixtures. Each plot consisted of two propagation trays with ‘Gelato-29’ cuttings located in every edge-adjacent cell (26 cells) in each tray. The remaining cells comprised the inner three rows (i.e., 3 × 8 cells) of the tray, into which cuttings of each of the three treatment cultivars were placed, with each cultivar randomly allocated to one row on a per-tray basis. With this arrangement, there were 16 subsamples per cultivar in each plot. The cuttings were misted with RO water on the foliage at the time of insertion (day 0) and then once on each of days 2, 4, and 6. On day 8, 0.8 L of nutrient solution was added to the bottom of each tray and topped up with nutrient solution thereafter, as needed. The nutrient solution was composed of Dutch Nutrients Gro A and Gro B (Homegrown Hydroponics, Toronto, ON, Canada), each at 5 mL·L⁻¹ in RO water, resulting in an EC of 1.7 dS·m⁻¹ and a pH of 5.8. On day 14, the cuttings were divided into two groups that were either harvested to evaluate rooting or transplanted and grown for an additional 21 d to evaluate transplant success (described later in this article).

Data loggers (HOBO MX2301A; Onset Computer Corporation, Bourne, MA, USA), positioned at fixture level in the center of each enclosure, were used to record temperature and RH on 60-s intervals throughout the propagation period. The daytime temperature and RH were 26 ± 3°C and 30% ± 8%, 26 ± 3°C and 30% ± 8%, and 25 ± 2°C and 30% ± 8% [mean ± standard deviation (SD), n = 14,400] for the three enclosures, respectively. The nighttime temperature and RH were 23 ± 2°C and 37% ± 5%, 23 ± 2°C and 36% ± 5%, and 23 ± 2°C and 36% ± 5% (mean ± SD, n = 7200) for the three enclosures, respectively. The inside of the propagation lids were wiped down with sterile cloths each time the cuttings were evaluated for rooting (described later in this article) to remove condensed water for pathogen suppression. Periodic point measurements of the aerial environment within the propagation domes were taken with a minia-

ture thermo-hygrometer (E100; Mondi Products, Vancouver, BC, Canada) (e.g., Fig. 1B, dome on the far left).

Spectrum treatments. The experiment was a randomized complete block design with six spectrum treatments and three blocks. All treatments had a daily photoperiod of 16 h (0600–2200 HR) and a target PPFD of 200 μmol·m⁻²·s⁻¹ at the surface of the propagation lids (Table 1). The spectrum, intensity, and uniformity were measured on a 0.1 × 0.1-m square grid (i.e., 25 locations), centered under the light fixtures to cover the space occupied by two propagation trays in each plot. Spectrum and intensity were measured using a radiometrically calibrated spectrometer (XR-Flame-S; Ocean Optics, Dunedin, FL, USA) coupled to a CCC cosine-corrector with a 1.9 m × 400 μm ultraviolet-Vis optical fiber. The PAR-Spec subroutine in SpectraSuite (Ocean Optics) was used for spectrum and intensity characterization that were measured at the beginning and end of the experiment.

Photon flux ratios of UVA (315–400 nm), blue (B, 400–500 nm), green (G, 500–600 nm), red (R, 600–700 nm), and far red (FR, 700–800 nm) were used to define the spectrum combinations in each treatment (Table 1). Within each treatment, only wavebands with photon flux integrals that comprise ≥ 0.5% of the total photon flux were used to define each respective treatment. Further, although UVA and FR wavebands are described in the spectrum treatments, wavelengths outside the photosynthetically active radiation (i.e., PAR; 400–700 nm) spectrum were excluded from the intensity (i.e.,
PPFD) measurements. Some treatments had fixed spectra throughout the entire propagation period, and others comprised dynamic spectral combinations that varied temporally throughout the propagation period. The treatments were as follows: 1) F, fixed fluorescent light spectrum (control); 2) BR, fixed spectrum of B15:R85; 3) B, B75:R25 on day 0–2 followed by B15:R85 on day 2–14; 4) B+UVA, B75:R25 on day 0–2 followed by B15:R85 on day 2–14 plus 15 μmol·m⁻²s⁻¹ of UVA on day 7–14; 5) B50, B15:R85 on day 0–7 followed by B50:R50 on day 7–14; and 6) W, fixed white spectrum (5700K). The initiation of the light spectrum treatments on day 0 and all subsequent spectrum changes occurred between 1000 and 1400 HR (i.e., during the middle portion of a given day’s photoperiod).

Several lighting technologies were used, alone or in combination, to provide the spectrum treatments. The control treatment used fluorescent lighting to match one of the most common propagation practices of commercial cultivators. The control plots each had two fluorescent tubes (F28W/T5/835/ECO; GE Lighting, East Cleveland, OH, USA). The spectral combinations in all other treatments were provided by LEDs. The UVA spectrum (custom-made LED bars; Yunustech, Mississauga, ON, Canada) had a peak wavelength and full-width at half-maximum intensity (FWHM) of 385 nm and 11 nm, respectively. The intensity of the UVA LEDs was modified using constant current dimmers. The remaining spectrum combinations were all provided by commercial LED bars (Toplight-Targeted Spectrum; Lumigrow, Emeryville, CA, USA), which contained three spectrum channels: blue, white, and red. The intensity of each channel was controlled with pulse-width modulation using software (smartPAR Horticulture Lighting Software Version 1.21.2; Lumigrow). The peak wavelengths and FWHM were 445 nm and 17 nm for blue and 665 nm and 16 nm for red channels, respectively. The white channel, which was composed of phosphor-converted blue LEDs, had two peaks with peak wavelengths and FWHM of 440 nm and 23 nm and 595 nm and 135 nm, respectively.

All lighting fixtures in each plot were hung 0.4 m apart (note, all plots, and centered over the plots. In the control age of growing media surface area that was observed by root tissue) were estimated in increments of 5% (i.e., 0% to 5%, 5% to 10%, 10% to 15%, etc.) and were recorded as root index (RI) values from 0 to 11 (Supplemental Table). The mean photon flux ratios, PPFD, within-plot PPFD uniformity (i.e., minimum PPFD/average PPFD), and phytochrome photostationary state [PPS; based on Sager et al. (1988)] were determined from raw spectral data for each treatment using a Microsoft Excel tool (Mah et al. 2019) and are presented in Table 1. The relative photon flux distribution of the different spectral combinations in each treatment (outlined in Table 1) are provided graphically in Supplemental Fig. 1.

Rooting and growth measurements. The cuttings were investigated regularly for the presence of visible roots. Each plug was investigated 3, 7, 9, 10, 13, and 14 d after insertion for root emergence on all exterior surfaces of each plug. The root coverage percentages (i.e., the percentage of growing media surface area that was obscured by root tissue) were estimated in increments of 5% (i.e., 0% to 5%, 5% to 10%, 10% to 15%, etc.) and were recorded as root index (RI) values from 0 to 11 (Supplemental Table).

Table 1. The photon flux ratios, photosynthetic photon density (PPFD; μmol·m⁻²·s⁻¹), uniformity, and phytochrome photostationary state (PPS) of the different spectrum treatments during the 15-day rooting phase of the cannabis cuttings.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Description</th>
<th>Day(s)</th>
<th>Photon flux ratio</th>
<th>PPFD (μmol·m⁻²·s⁻¹)</th>
<th>Uniformity</th>
<th>PPS</th>
</tr>
</thead>
<tbody>
<tr>
<td>F</td>
<td>Fluorescent (6500 K CCT) spectrum</td>
<td>0–14</td>
<td>1 29 41 25 4</td>
<td>203 ± 22</td>
<td>0.80</td>
<td>0.8</td>
</tr>
<tr>
<td>BR</td>
<td>Ratio of blue (B) and red (R) spectra of B15:R85</td>
<td>0–14</td>
<td>– 15 – 85 –</td>
<td>205 ± 12</td>
<td>0.89</td>
<td>0.88</td>
</tr>
<tr>
<td>B</td>
<td>High B on day 0–2 followed by the BR</td>
<td>0–2</td>
<td>75 – 25 –</td>
<td>203 ± 10</td>
<td>0.92</td>
<td>0.80</td>
</tr>
<tr>
<td></td>
<td>spectrum on day 2–14</td>
<td>2–14</td>
<td>– 15 – 85</td>
<td>204 ± 10</td>
<td>0.91</td>
<td>0.88</td>
</tr>
<tr>
<td>B+UVA</td>
<td>High B on day 0–2, followed by BR</td>
<td>0–2</td>
<td>– 75 – 25 –</td>
<td>206 ± 12</td>
<td>0.87</td>
<td>0.80</td>
</tr>
<tr>
<td></td>
<td>spectrum on day 2–14, plus 15 μmol·m⁻²·s⁻¹ of UVA on day 7–14</td>
<td>2–7</td>
<td>– 15 – 85 –</td>
<td>205 ± 10</td>
<td>0.89</td>
<td>0.88</td>
</tr>
<tr>
<td></td>
<td>of UVA on day 7–14</td>
<td>7–14</td>
<td>7 14 – 79</td>
<td>205 ± 10</td>
<td>0.64</td>
<td>0.87</td>
</tr>
<tr>
<td>B50</td>
<td>The BR spectrum on day 0–7 followed by equal amounts of B and R on day 7–14</td>
<td>0–7</td>
<td>– 15 – 85 –</td>
<td>202 ± 10</td>
<td>0.88</td>
<td>0.88</td>
</tr>
<tr>
<td></td>
<td>White (5700 K CCT) LED spectrum</td>
<td>0–14</td>
<td>23 48 26 3</td>
<td>200 ± 14</td>
<td>0.86</td>
<td>0.84</td>
</tr>
</tbody>
</table>

¹ Photon flux ratios of ultraviolet-A (UVA, 315–400 nm), blue (B, 400–500 nm), green (G, 500–600 nm), red (R, 600–700 nm), and far red (FR, 700–800 nm).
² PPFD are means ± standard deviation (n = 3) from the three replicate plots.
³ Uniformity was calculated, on a per-plot basis, as the mean of minimum PPFD/average PPFD across the replications (n = 3).
⁴ Phytochrome photostationary state, calculated from Sager et al. (1988).
⁵ Correlated color temperature.
1) based on the rooting stage parameter described in Birlanga et al. (2015). Logistical constraints prevented the (necessarily destructive) assessments (i.e., prising apart the rockwool plug) of root initiation. After the 14 d of spectrum treatments, all cuttings were evaluated for an array of rooting and aboveground attributes. The RI on day 14 was used to indicate the root density of each cutting at the time of transplant, as well as the overall percentage of cuttings from each treatment × cultivar combination that had developed visible roots (i.e., RI > 0). Chlorophyll content on the youngest fully expanded leaf of each cutting was measured using a chlorophyll meter (SPAD 502; Spectrum Technologies Inc., Aurora, IL, USA). Stem thickness (ST) was measured at the first internode of each cutting using a digital caliper. The presence of new aboveground growth was recorded for each cutting that had new leaves or nodes.

For each cultivar × tray combination, two plants that had RI > 0 were randomly selected and transplanted into rockwool blocks (0.15 × 0.15 × 0.15 m; Grodan) that were presoaked with saturation with rainwater and grown vegetatively (16-h photoperiod) under 277 ± 6 μmol·m−2·s−1 (mean ± SD, n = 6) from metal halide lamps (MHD NXTLP RST MH; PL Light Systems Inc., Beamsville, ON, Canada) for 21 d. The initial height, measured from growing media surface to the highest point on the plant, and the number of nodes of each plant were recorded. Plants were randomly arranged 0.24 m apart on a bench and fertigated using a solution composed of Dutch Nutrients Gro A and Gro B (Homegrown Hydroponics), each added at 5 mL·L−1 to rainwater (EC < 0.1 dS·m−1), with a resulting EC of ≈1.8 dS·m−1 and pH of ≈5.7. The rainwater consistently had an EC ≤ 0.1 dS·m−1 and was treated with 100 ppm hydrogen peroxide at least 24 h before use. The transplants were not fertigated for the first 3 d to encourage root growth, then were drip-irrigated twice daily at 2 L·h−1 for 540 s, such that each plant received ≈0.6 L·d−1.

The remaining six cuttings from each cultivar × tray combination were destructively harvested by cutting the stems at growing media level and oven-drying the root balls (i.e., rockwool plug with roots in individual paper bags) to constant weight at 65 °C (Isotemp Oven Model 655G; Fisher Scientific, East Lynne, CT, USA). Dry weights (DW) were measured using an analytical balance (Mettler Toledo AE 100; Mettler Toledo, Columbus, OH, USA). Since the roots could not be extracted from the rockwool plugs, the combined DW of the rockwool and roots (i.e., root ball) were converted into relative values [relative root dry weight of plugs (RRDWP)] using the formula: root ball DW/DW of the heaviest root ball × 100%.

Chlorophyll content was measured thrice, then averaged, on the youngest fully expanded leaf using a chlorophyll meter (SPAD 502). The total aboveground fresh weight (FW) of each plant was recorded (AX622N/E Adventure Precision Balance; OHAUS Corporation, Parsippany, NJ, USA) and then dried at 65 °C (Isotemp Oven Model 655G) to constant weight to obtain aboveground DW. Aboveground water content (WC) was calculated using the following equation: [(FW − DW)/FW] × 100%. The rockwool blocks from the transplants were dried at 65 °C to constant weight and then the combined DW of the rockwool and roots (i.e., root ball) were converted into relative values [relative root dry weight of transplants (RRDWT)] using the following formula: root ball DW/DW of the heaviest root ball.

**Statistical analysis.** Data were analyzed using generalized linear mixed models using SAS (Statistical Analysis Software University Edition; SAS Institute Inc., Cary, NC, USA). For each growth attribute, spectrum (S), cultivar (C), and the interaction between spectrum and cultivar (S × C) are followed by NS or * to denote that treatment effects are not significant or are significant at P ≤ 0.05, respectively.

<table>
<thead>
<tr>
<th>Treatment1</th>
<th>Rooted cuttings (%)</th>
<th>Root index</th>
<th>Relative root dry wt2</th>
<th>SPAD value</th>
<th>Stem thickness (mm)</th>
<th>New aboveground growth (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>‘Gelato-27’</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>96 ± 2.2 a</td>
<td>3.2 ± 0.28 a</td>
<td>0.99 ± 0.031 a</td>
<td>52 ± 0.6 a</td>
<td>4.2 ± 0.10 b</td>
<td>92 ± 5.5 a</td>
</tr>
<tr>
<td>BR</td>
<td>100 ± 0.0 a</td>
<td>4.1 ± 0.09 a</td>
<td>1.0 ± 0.008 a</td>
<td>51 ± 0.8 a</td>
<td>4.3 ± 0.05 ab</td>
<td>98 ± 2.1 a</td>
</tr>
<tr>
<td>B</td>
<td>98 ± 2.2 a</td>
<td>4.1 ± 0.32 a</td>
<td>1.0 ± 0.062 a</td>
<td>51 ± 1.4 a</td>
<td>4.7 ± 0.09 a</td>
<td>90 ± 1.5 a</td>
</tr>
<tr>
<td>B+UVA</td>
<td>98 ± 2.1 a</td>
<td>3.9 ± 0.10 a</td>
<td>1.0 ± 0.041 a</td>
<td>52 ± 1.1 a</td>
<td>4.4 ± 0.19 ab</td>
<td>98 ± 2.1 a</td>
</tr>
<tr>
<td>B50</td>
<td>98 ± 2.1 a</td>
<td>3.4 ± 0.20 a</td>
<td>0.98 ± 0.013 a</td>
<td>51 ± 0.0 a</td>
<td>4.4 ± 0.16 ab</td>
<td>94 ± 3.6 a</td>
</tr>
<tr>
<td>W</td>
<td>98 ± 2.1 a</td>
<td>4.1 ± 0.22 a</td>
<td>0.95 ± 0.029 a</td>
<td>52 ± 1.0 a</td>
<td>4.4 ± 0.08 ab</td>
<td>100 ± 0.0 a</td>
</tr>
<tr>
<td>Mean3</td>
<td>98 ± 0.7 A</td>
<td>3.8 ± 0.11 A</td>
<td>0.99 ± 0.013 A</td>
<td>51 ± 0.4 B</td>
<td>4.4 ± 0.06 A</td>
<td>97 ± 1.3 A</td>
</tr>
<tr>
<td><strong>‘Grace’</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>94 ± 3.6 a</td>
<td>3.6 ± 0.14 a</td>
<td>0.97 ± 0.019 ab</td>
<td>55 ± 0.1 a</td>
<td>3.7 ± 0.25 a</td>
<td>88 ± 3.6 a</td>
</tr>
<tr>
<td>BR</td>
<td>92 ± 4.2 a</td>
<td>3.4 ± 0.12 a</td>
<td>0.95 ± 0.033 ab</td>
<td>52 ± 1.4 a</td>
<td>3.4 ± 0.28 a</td>
<td>98 ± 2.1 a</td>
</tr>
<tr>
<td>B</td>
<td>100 ± 0.0 a</td>
<td>3.1 ± 0.19 a</td>
<td>0.97 ± 0.005 ab</td>
<td>53 ± 1.4 a</td>
<td>3.6 ± 0.24 a</td>
<td>96 ± 2.1 a</td>
</tr>
<tr>
<td>B+UVA</td>
<td>91 ± 4.3 a</td>
<td>4.0 ± 0.22 a</td>
<td>1.0 ± 0.035 a</td>
<td>53 ± 1.0 a</td>
<td>3.6 ± 0.17 a</td>
<td>94 ± 3.6 a</td>
</tr>
<tr>
<td>B50</td>
<td>96 ± 2.1 a</td>
<td>3.8 ± 0.35 a</td>
<td>0.97 ± 0.030 ab</td>
<td>53 ± 0.8 a</td>
<td>3.5 ± 0.20 a</td>
<td>90 ± 5.5 a</td>
</tr>
<tr>
<td>W</td>
<td>94 ± 6.3 a</td>
<td>3.4 ± 0.38 a</td>
<td>0.93 ± 0.032 b</td>
<td>52 ± 0.2 a</td>
<td>3.4 ± 0.05 a</td>
<td>96 ± 4.4 a</td>
</tr>
<tr>
<td>Mean</td>
<td>94 ± 1.5 A</td>
<td>3.5 ± 0.11 A</td>
<td>0.98 ± 0.011 A</td>
<td>53 ± 0.4 A</td>
<td>3.5 ± 0.08 C</td>
<td>93 ± 1.6 AB</td>
</tr>
<tr>
<td><strong>‘Meridian’</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>94 ± 3.6 a</td>
<td>3.2 ± 0.62 a</td>
<td>0.98 ± 0.030 a</td>
<td>49 ± 1.3 a</td>
<td>4.0 ± 0.14 a</td>
<td>71 ± 11.5 a</td>
</tr>
<tr>
<td>BR</td>
<td>94 ± 3.6 a</td>
<td>3.9 ± 0.49 a</td>
<td>0.97 ± 0.010 a</td>
<td>49 ± 0.7 a</td>
<td>4.0 ± 0.09 a</td>
<td>94 ± 3.6 a</td>
</tr>
<tr>
<td>B</td>
<td>100 ± 0.0 a</td>
<td>3.4 ± 0.22 a</td>
<td>0.97 ± 0.011 a</td>
<td>48 ± 1.8 a</td>
<td>4.2 ± 0.18 a</td>
<td>96 ± 2.1 a</td>
</tr>
<tr>
<td>B+UVA</td>
<td>96 ± 2.2 a</td>
<td>4.1 ± 0.14 a</td>
<td>0.97 ± 0.011 a</td>
<td>50 ± 1.0 a</td>
<td>4.2 ± 0.07 a</td>
<td>89 ± 5.6 a</td>
</tr>
<tr>
<td>B50</td>
<td>100 ± 0.0 a</td>
<td>3.5 ± 0.17 a</td>
<td>1.0 ± 0.023 a</td>
<td>50 ± 0.2 a</td>
<td>4.0 ± 0.09 a</td>
<td>92 ± 4.2 a</td>
</tr>
<tr>
<td>W</td>
<td>98 ± 2.1 a</td>
<td>3.3 ± 0.39 a</td>
<td>0.95 ± 0.047 a</td>
<td>50 ± 1.0 a</td>
<td>3.9 ± 0.05 a</td>
<td>98 ± 2.1 a</td>
</tr>
<tr>
<td>Mean</td>
<td>97 ± 1.1 A</td>
<td>3.6 ± 0.15 A</td>
<td>1.0 ± 0.010 A</td>
<td>50 ± 0.4 C</td>
<td>4.0 ± 0.05 B</td>
<td>90 ± 3.0 B</td>
</tr>
</tbody>
</table>

| S | NS | * | NS | NS | NS |
| C | NS | NS | NS | NS | NS |
| S × C | NS | NS | NS | NS | NS |

1 Spectrum treatments are described in Table 1 and spectrum distributions are provided graphically in Supplemental Fig. 1.

2 Relative root dry weight of the plugs (RRDWp) was calculated as: root ball DW/DW of the heaviest root ball.

3 Overall intracultivar treatment means ± standard error (SE) (n = 3). Any treatment means within a column for a given cultivar followed by the same lowercase letter are not significantly different at P ≤ 0.05, according to the Tukey-Kramer multiple comparisons test.

4 Overall cultivar means ± SE (n = 18). Any overall cultivar means within a column followed by the same uppercase letter are not significantly different at P ≤ 0.05, according to the Tukey-Kramer multiple comparisons test.

5 For each growth attribute, spectrum (S), cultivar (C), and the interaction between spectrum and cultivar (S × C) are followed by NS or * to denote that treatment effects are not significant or are significant at P ≤ 0.05, respectively.
Data were subjected to an analysis of variance, followed by Tukey-Kramer’s tests to determine differences among spectrum treatments, cultivars, and spectrum × cultivar combinations at a significance level of $P \leq 0.05$. When intracultivar spectrum treatment, interculivar spectrum treatment, cultivar, and overall experiment means are presented, data are presented as means ± standard error (SE), where $n = 3, 9, 18$, and 54, respectively.

**Results**

*Rooting.* There were no obvious signs of rooting on the outer surfaces of the plugs until 9 d after the start of the experiment. Despite this time lag, there was a high percentage of rooted cuttings (i.e., RI > 0) at day 14 (Table 2), with an overall experiment mean of 96% ± 0.7%. Although there were no intracultivar spectrum treatment effects on RI, when the data from the three cultivars were combined, the RI was 21% higher in the B+UVA vs. F spectrum treatments (Fig. 2A). In the harvested cuttings, the RRDWP in ‘Gelato-27’ was 8% higher in the B+UVA vs. W treatments (Table 2). Further, when the data from the three cultivars were combined, the RRDWP in the B+UVA, B50, F, and B treatments were 4% higher than the W treatment (Fig. 2B).

*Aboveground growth of rooted cuttings.* Cuttings were evaluated at the end of the 15-day propagation stage to characterize their development before the transition into the vegetative stage. There were no spectrum effects on SPAD value (i.e., a leaf area-based index of chlorophyll content) with an overall mean of 51 ± 0.3; however, the SPAD value in ‘Grace’ was 4% and 6% higher than in ‘Gelato-27’ and ‘Meridian’, respectively, whereas ‘Gelato-27’ had 2% higher SPAD value than ‘Meridian’ (Table 2). ‘Gelato-27’ cuttings had 12% higher ST in the B vs. F treatments (Table 2). Further, across all spectrum treatments, ‘Gelato-27’ stems were 10% and 26% thicker than ‘Meridian’ and ‘Grace’, respectively, and ‘Meridian’ had 14% thicker stems than ‘Grace’ (Table 2). Across all cultivars, the cuttings in the B treatment had 8% thicker stems vs. both BR and W treatments (Fig. 2C). There were no spectrum effects on new aboveground growth within each cultivar, but ‘Gelato-27’ had 8% more cuttings exhibiting new aboveground growth vs. ‘Meridian’ (Table 2). Across all cultivars, 16% more cuttings had new aboveground growth under the W, B, BR, and B+UVA treatments vs. the F treatment (Fig. 2D).

*Discussion*

This trial was designed to evaluate the effects of light spectrum on the rooting, growth, and transplant success of clonal cuttings from different cannabis cultivars that reportedly had varying rooting rates. Traditional fluorescent lighting along with five LED spectra, composed of two fixed-spectrum (W and BR) and three dynamic-spectrum (B, B+UVA, and B50) treatments, were investigated. Some of the spectrum treatment selections were based on previous findings in the literature that high proportions of B can promote root initiation (Gil et al. 2020) and make transplants more compact (Ying et al. 2020), whereas UVA can promote root growth and

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*Fig. 2.* Root index (A), root dry weight of plugs (RRDWP) relative to the heaviest root ball (B), stem thickness (C), and percentage of cuttings that exhibited new aboveground growth (D) for cuttings of three cannabis cultivars rooted under different spectrum treatments for 15 d. 1) F, fixed fluorescent light spectrum (control); 2) BR, fixed spectrum of B15:R85; 3) B, B75:R25 on day 0–2 followed by B15:R85 on day 2–14; 4) B+UVA, B75:R25 on day 0–2 followed by B15:R85 on day 2–14 plus 15 μmol·m⁻²·s⁻¹ of UVA on day 7–14; 5) B50, B15:R85 on day 0–7 followed by B50:R50 on day 7–14; and 6) W, fixed white spectrum (5700K). The photosynthetic photon flux density (PPFD; 400–700 nm) was ≈200 μmol·m⁻²·s⁻¹ for all treatments. Bars represent means ± standard error of all three cultivars combined ($n = 9$). Bars that share the same lowercase letter are not significantly different at $P \leq 0.05$, according to Tukey-Kramer multiple comparisons test.
Table 3. The effects of spectrum and cultivar on increase in height (ΔH), increase in number of nodes (ΔNN), internode length (IL), stem thickness (ST), SPAD value (i.e., chlorophyll content index), aboveground fresh weight (FW), aboveground dry weight (DW), aboveground water content (WC), and relative root dry weight (RRDWT) for ‘Gelato’, ‘Grace’, and ‘Meridian’ cannabis transplants, after the 15-day spectrum treatment period (±200 μmol·m−2·s−1), then grown for 21 d in a vegetative environment under a metal halide spectrum (±275 μmol·m−2·s−1).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Increase in HT (cm)</th>
<th>Increase in no. of nodes</th>
<th>Internode length (cm)</th>
<th>Stem thickness (mm)</th>
<th>SPAD value</th>
<th>Above ground FW (g)</th>
<th>Above ground DW (g)</th>
<th>Water content (%)</th>
<th>Relative root dry wt (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>'Gelato'</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>38 ± 2.0 a</td>
<td>10.2 ± 0.22 a</td>
<td>3.8 ± 0.25 a</td>
<td>8.1 ± 0.23 a</td>
<td>55 ± 0.9</td>
<td>91 ± 10.4 a</td>
<td>20 ± 2.7 a</td>
<td>79 ± 0.5 a</td>
<td>0.98 ± 0.006 a</td>
</tr>
<tr>
<td>BR</td>
<td>40 ± 2.0 a</td>
<td>10.8 ± 0.46 a</td>
<td>3.8 ± 0.30 a</td>
<td>8.4 ± 0.28 a</td>
<td>54 ± 0.9</td>
<td>101 ± 6.6 a</td>
<td>22 ± 1.3 a</td>
<td>78 ± 0.5 a</td>
<td>0.98 ± 0.016 a</td>
</tr>
<tr>
<td>B</td>
<td>39 ± 1.3 a</td>
<td>10.4 ± 0.22 a</td>
<td>3.9 ± 0.17 a</td>
<td>8.2 ± 0.09 a</td>
<td>54 ± 2.0</td>
<td>92 ± 4.7 a</td>
<td>20 ± 1.1 a</td>
<td>79 ± 0.2 a</td>
<td>0.98 ± 0.010 a</td>
</tr>
<tr>
<td>B+UVA</td>
<td>40 ± 0.9 a</td>
<td>10.8 ± 0.29 a</td>
<td>3.8 ± 0.19 a</td>
<td>8.6 ± 0.03 a</td>
<td>55 ± 1.3</td>
<td>108 ± 3.2 a</td>
<td>24 ± 0.9 a</td>
<td>77 ± 0.4 a</td>
<td>1.00 ± 0.009 a</td>
</tr>
<tr>
<td>B50</td>
<td>37 ± 2.6 a</td>
<td>11.2 ± 0.17 a</td>
<td>3.6 ± 0.04 a</td>
<td>8.1 ± 0.11 a</td>
<td>55 ± 0.7</td>
<td>99 ± 4.5 a</td>
<td>22 ± 1.0 a</td>
<td>78 ± 0.2 a</td>
<td>0.98 ± 0.003 a</td>
</tr>
<tr>
<td>W</td>
<td>39 ± 1.3 a</td>
<td>10.8 ± 0.43 a</td>
<td>3.7 ± 0.21 a</td>
<td>8.3 ± 0.17 a</td>
<td>54 ± 1.3</td>
<td>96 ± 8.0 a</td>
<td>21 ± 1.7 a</td>
<td>78 ± 0.3 a</td>
<td>0.99 ± 0.004 a</td>
</tr>
<tr>
<td>Mean</td>
<td>39 ± 0.5 C</td>
<td>10.7 ± 0.13 A</td>
<td>3.8 ± 0.07 B</td>
<td>8.3 ± 0.07 C</td>
<td>55 ± 0.5</td>
<td>97 ± 2.7 A</td>
<td>21 ± 0.7 A</td>
<td>78 ± 0.2 A</td>
<td>1.00 ± 0.004 A</td>
</tr>
<tr>
<td>'Grace'</td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>43 ± 1.9 a</td>
<td>8.7 ± 0.36 a</td>
<td>5.1 ± 0.41 a</td>
<td>8.7 ± 0.19 a</td>
<td>58 ± 0.6</td>
<td>89 ± 5.7 a</td>
<td>19 ± 1.1 a</td>
<td>79 ± 0.4 a</td>
<td>0.99 ± 0.005 a</td>
</tr>
<tr>
<td>BR</td>
<td>43 ± 2.0 a</td>
<td>9.3 ± 0.00 a</td>
<td>4.7 ± 0.09 a</td>
<td>8.7 ± 0.16 a</td>
<td>56 ± 1.4</td>
<td>97 ± 3.2 a</td>
<td>20 ± 0.7 a</td>
<td>79 ± 0.1 a</td>
<td>1.00 ± 0.009 a</td>
</tr>
<tr>
<td>B</td>
<td>38 ± 1.7 a</td>
<td>9.3 ± 0.22 a</td>
<td>4.1 ± 0.22 a</td>
<td>8.2 ± 0.33 a</td>
<td>55 ± 1.4</td>
<td>88 ± 5.1 a</td>
<td>19 ± 1.1 a</td>
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<td>0.97 ± 0.002 a</td>
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<td>8.8 ± 0.09 a</td>
<td>59 ± 0.4</td>
<td>100 ± 0.7 a</td>
<td>22 ± 0.04 a</td>
<td>78 ± 0.2 a</td>
<td>0.98 ± 0.004 a</td>
</tr>
<tr>
<td>B50</td>
<td>42 ± 0.9 a</td>
<td>9.8 ± 0.08 a</td>
<td>4.4 ± 0.09 a</td>
<td>8.9 ± 0.10 a</td>
<td>57 ± 0.5</td>
<td>97 ± 2.7 a</td>
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<tr>
<td>W</td>
<td>44 ± 0.9 a</td>
<td>9.9 ± 0.22 a</td>
<td>4.4 ± 0.10 a</td>
<td>8.8 ± 0.21 a</td>
<td>58 ± 0.7</td>
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<td>79 ± 0.3 a</td>
<td>0.97 ± 0.005 a</td>
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<tr>
<td>Mean</td>
<td>42 ± 0.6 B</td>
<td>9.4 ± 0.13 B</td>
<td>4.6 ± 0.10 A</td>
<td>8.7 ± 0.09 B</td>
<td>57 ± 0.5</td>
<td>95 ± 1.7 A</td>
<td>20 ± 0.4 A</td>
<td>79 ± 0.2 A</td>
<td>0.99 ± 0.003 A</td>
</tr>
<tr>
<td>'Meridian'</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>45 ± 1.7 a</td>
<td>9.5 ± 0.55 a</td>
<td>4.7 ± 0.21 a</td>
<td>9.2 ± 0.23 a</td>
<td>53 ± 0.5</td>
<td>94 ± 3.8 a</td>
<td>21 ± 0.7 a</td>
<td>78 ± 0.3 a</td>
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</tr>
<tr>
<td>BR</td>
<td>47 ± 0.4 a</td>
<td>10.4 ± 0.61 a</td>
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<td>9.2 ± 0.20 a</td>
<td>56 ± 1.1</td>
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<td>21 ± 0.4 a</td>
<td>78 ± 0.3 a</td>
<td>1.00 ± 0.009 a</td>
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<td>B</td>
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<td>4.8 ± 0.31 a</td>
<td>9.3 ± 0.15 a</td>
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<td>B+UVA</td>
<td>45 ± 1.1 a</td>
<td>10.6 ± 0.30 a</td>
<td>4.3 ± 0.22 a</td>
<td>8.9 ± 0.06 a</td>
<td>56 ± 0.4</td>
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<tr>
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<td>55 ± 1.3</td>
<td>102 ± 2.2 a</td>
<td>23 ± 1.0 a</td>
<td>77 ± 0.6 a</td>
<td>1.00 ± 0.005 a</td>
</tr>
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<td>46 ± 1.2 a</td>
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<td>4.9 ± 0.20 a</td>
<td>8.7 ± 0.65 a</td>
<td>54 ± 0.5</td>
<td>89 ± 8.0 a</td>
<td>19 ± 1.8 a</td>
<td>79 ± 0.2 a</td>
<td>0.98 ± 0.008 a</td>
</tr>
<tr>
<td>Mean</td>
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<td>4.7 ± 0.10 A</td>
<td>9.0 ± 0.12 A</td>
<td>54 ± 0.4</td>
<td>96 ± 2.0 A</td>
<td>21 ± 0.5 a</td>
<td>78 ± 0.3 A</td>
<td>1.00 ± 0.004 A</td>
</tr>
</tbody>
</table>

Spectrum treatments are described in Table 1 and spectrum images are provided graphically in Supplemental Fig. 1. Relative root dry weight of the transplants (RRDWT) was calculated as: root ball DW/DW of the heaviest root ball. Overall intracultivar treatment means ± standard error (SE) (n = 3). Any treatment means within a column for a given cultivar followed by the same lowercase letter are not significantly different at P ≤ 0.05, according to the Tukey-Kramer multiple comparisons test. Overall cultivar means ± SE (n = 18). Any overall cultivar means within a column followed by the same uppercase letter are not significantly different at P ≤ 0.05, according to the Tukey-Kramer multiple comparisons test. For each growth attribute, spectrum (S), cultivar (C), and the interaction between spectrum and cultivar (S × C) are followed by NS or * to denote that treatment effects are not significant or are significant at P ≤ 0.05, respectively.
energy efficiency and color rendering, some modern horticultural LED technologies contain a limited proportion of W LEDs to promote improved color rendering while still maintaining the relatively high efficacy emblematic of fixtures composed predominantly of B and R LEDs. Accordingly, the reported efficacies are normally higher than fixtures composed predominantly of W LEDs and lower than fixtures composed entirely of B and R LEDs (DesignLights Consortium 2021). For a given production system, the optimum LED spectrum distribution for cannabis production will depend on the growth stage and cultivators’ specific production goals. In the relatively short propagation stage, the energy efficiency benefits of spectra with high proportions of narrowband R and B may be largely offset by low color rendering (Park and Runkle 2018) and reduced occupant comfort largely offset by low color rendering (Park and Runkle 2018) and reduced occupant comfort.


Supplemental Fig. 1. Relative spectral photon flux distribution for cannabis cuttings rooted under the different spectrum treatments for 15 days: 1) F, fixed fluorescent light spectrum (control); 2) BR, fixed spectrum of B15:R85; 3) B, B75:R25 on day 0–2 followed by B15:R85 on day 2–14; 4) B+UVA, B75:R25 on day 0–2 followed by B15:R85 on day 2–14 plus 15 μmol m⁻² s⁻¹ of UVA on day 7–14; 5) B50, B15:R85 on day 0–7 followed by B50:R50 on day 7–14; and 6) W, fixed white spectrum (5700K).
Supplemental Table 1. The conversion from estimated root coverage percentages observed on all surfaces of the rockwool plug to root index (RI).

<table>
<thead>
<tr>
<th>Estimated root coverage range (%)</th>
<th>Root index</th>
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</tr>
<tr>
<td>0–5</td>
<td>1</td>
</tr>
<tr>
<td>5–10</td>
<td>2</td>
</tr>
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