



Communication The Biometric Parameters of Microgreen Crops Grown under Various Light Conditions

Barbara Frąszczak^{1,*} and Monika Kula-Maximenko²

- ¹ Department of Vegetable Crops, Poznań University of Life Sciences, ul. Dąbrowskiego 159, 60-594 Poznań, Poland
- ² The Franciszek Górski Institute of Plant Physiology, Polish Academy of Sciences, ul. Niezapominajek 21, 30-239 Kraków, Poland; m.kula@ifr-pan.edu.pl
- * Correspondence: barbara.fraszczak@up.poznan.pl; Tel.: + 48-61-846-6320

Abstract: Microgreens are becoming increasingly popular both as horticultural crops and as vegetables consumed by humans. They are classified as foods of high nutritional value. Twenty-eight microgreens crops were grown in a growth chamber under fully controlled conditions in order to determine how different light treatments affected their growth rate. The plants were grown under three light sources emitting red/blue ratios of about 6.7, 0.6, and 1.6 units (Red light, Blue light, and R + B light, respectively). Apart from that, the spectrum contained 10% yellow and orange light and 10% green light. The fresh weight of the plants ranged from 8 (perilla) to 1052 mg (nasturtium), whereas the length ranged for the same plants from 2.0 to 26.2 cm. The nasturtium was particularly strongly distinguished from the other species by the high values of its biometric parameters. The fresh mass of most of the other microgreens ranged from 20 to 100 mg, whereas their height ranged from 5 to 8 cm. Red light caused a significant increase in the fresh and dry weights of more than half of the species. The light spectrum had a lesser influence on the length of the plants. The research results showed considerable differences in the dynamics of growth of commonly cultivated microgreens.

Keywords: LEDs; red light; blue light; functional food

1. Introduction

Scientific publications define microgreens as tender, immature greens produced from the seeds of vegetables, herbs, and cereals, which have two fully developed cotyledons with or without the first two true leaves. Over the past few years, they have gained popularity as a new culinary trend. Although microgreens are small, they have surprisingly intense flavours, vivid colours, a crunchy texture, and they can be served as a new ingredient or as an edible addition to salads [1].

Depending on the cultivation system, weather conditions, and genotype, the duration of the growth cycle of microgreens may range from 7 to 28 days after emergence. Microgreens can be harvested after the appearance of the first true leaves when the cotyledons are fully developed and still firm, and, depending on the species, their length ranges from 10 to 20 cm [2] (pp. 403–432).

Microgreens can be grown in various environments, e.g., outdoors, in a greenhouse, and indoors. However, they are usually grown in a controlled environment with artificial lighting [3,4]. Nowadays, LED modules are considered the best option for indoor and vertical farming [5]. The authors of earlier studies usually used a light intensity of 100–300 μ mol m⁻² s⁻¹ PPFD to grow microgreens [6,7]. According to some authors, less than 100 μ mol m⁻² s⁻¹ is enough for microgreens to grow in an artificial light environment [8,9]. Both red and blue light effectively promote plant growth because they are better absorbed by photosynthetic pigments than other ranges of the light spectrum. Red and/or blue LEDs are most commonly used for growing microgreens because these spectra can increase the fresh weight of plants [10,11]. However, the ratio of red to blue light



Citation: Frąszczak, B.; Kula-Maximenko, M. The Biometric Parameters of Microgreen Crops Grown under Various Light Conditions. *Agriculture* **2022**, *12*, 576. https://doi.org/10.3390/ agriculture12050576

Academic Editors: Viktorija Vaštakaitė-Kairienė and Giedrė Samuolienė

Received: 25 March 2022 Accepted: 18 April 2022 Published: 20 April 2022

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Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). must be carefully selected because the spectra with larger blue fractions resulted in lower yields [12,13]. Fresh weight is one of the most important parameters used for the assessment of microgreens' growth. Crop yield is important for microgreen growers because it directly affects economic return [14]. The effect of light on fresh weight varies depending on the applied light spectra and species [8].

There are numerous scientific publications about microgreens. However, their authors usually focused on one or a few crops [15] or they assessed groups of active compounds (vitamins and minerals) [1,16]. It is known that there is a large number of plant species that can be eaten as microgreens [8,17]. However, there have been no studies comparing the dynamics of the growth of a wide range of microgreens under different light conditions. The aim of our preliminary research was to determine the growth of 28 microgreen crops under different light conditions.

2. Materials and Methods

2.1. Plant Material and Growth Conditions

The growth rate of 28 microgreen species and cultivars belonging to the following families was assessed: *Alliaceae* (Welsh onion), *Amaranthaceae* (garnet amaranth), *Apiaceae* (coriander, dill), *Asteraceae* (leaf chicory and lettuce), *Boraginaceae* (borage), *Brassicaceae* (black radish, broccoli, garden cress, green and red kale, purple kohlrabi, komatsuna, mibuna, Indian mustard, mizuna, red cabbage, red radish, rocket, watercress), *Chenopodiaceae* (red Swiss chard, spinach), *Fabaceae* (alfalfa, pea), *Lamiaceae* (green, purple, and lemon basil, perilla) and *Tropaeolaceae* (nasturtium). The common and scientific names, the botanical family, and the cultivar of these plants are listed in Supplementary Table S1.

All microgreens were grown in a growth chamber equipped with three tables, each of which had its own independent lighting (Figure S1). A different light spectrum composition was used on each table. All microgreen crops were sown in two special plastic trays (14 crops in each tray; $28 \times 54 \times 10$ cm) containing a peat substrate intended for the production of vegetable seedlings (Klasmann-Deilmann, Germany). The average amounts of nutrients (mg L⁻¹) in the substrate were as follows: N-NH₄–28; N-NO₃–252; P–87; K–278; Ca–579; Mg–294; S-SO₄–182; Na–37; Cl–13; pH in H₂O–5.49; EC–0.829 mS·cm⁻¹. Six containers, i.e., three replicates, were used in each experimental treatment. The substrate was moistened before sowing the seeds. Thirty seeds of each species were sown as densely as possible. The seeds were covered with a thin layer of sand and sprinkled. Subsequently, a lid was placed on top of the containers and the trays remained in darkness at 25 °C until germination (3 days). The average germination percentage was 98% and the mean germination time was 3 days.

During vegetation, day/night temperatures of $22/17 \pm 2$ °C were established with a 16-h photoperiod and relative humidity of 65–75%. The microgreens were grown under red, blue, green, and yellow light emitting diode (LED) panel units (Xlamp, type SMD, Seoul Semiconductor, South Korea), with an emission wavelength of 400–700 nm. The plants were grown in three light combinations differing in the red to blue light ratio (combination R–6.7, combination B–0.6, combination B + R–1.6). Red light accounted approximately for 3/4, 1/4, and 2/4 of the entire spectrum, respectively. A detailed description of the light sources is shown in Figure 1.

The LED panel was arranged in the growth chamber so as to ensure a homogeneous photosynthetic photon flux density (PPFD) of $130 \pm 4 \mu mol m^{-2} s^{-1}$ over the entire surface of the table. The PPFD was measured with a PAR-10 quantum sensor (Sonopan, Białystok, Poland). The spectral distribution of light treatments was measured with a BLACK-Comet CXR UV-VIS spectroradiometer (280–900 nm, StellarNet Inc., Tampa, FL, USA). The trays with the substrate were arranged randomly and systematically rotated every second day to enhance the uniformity of the light environment.





2.2. Measurement and Collection of Growth Parameter Data

At harvest time, i.e., 14 days after germination, the following morphological parameters were measured: fresh and dry weight of seedlings (mg, both parameters were measured using a laboratory scale WTB 200, r: 0.001 g., Radwag, Radom, Poland), and seedling length (including leaves, cm, using a ruler). The length, fresh mass, and dry weight were measured on 10 seedlings, which were randomly selected within each tray. The dry weight was calculated by drying the material to a constant weight at 105 °C for 24 h. The plant height/dry weight ratio (H/DW, cm g⁻¹) was also calculated.

2.3. Experiment Design and Statistical Analysis

The results were the means of the three cycles. The data were analysed with ANOVA. Differences between the means were estimated with Tukey's HSD test at a significance level of α = 0.05. The data were analysed statistically with the OriginPro 2020 program (OriginLab, USA).

Principal component analysis (PCA) was applied to find correlations between the measured parameters and the spectral composition of light. Hierarchical cluster analysis (HCA) was used to find similarities and differences between the species of microgreens, depending on the spectral composition of light and biometric parameters. The Euclidean distance was used in the HCA. The distance between similar groups was measured by means of the Ward algorithm. The OriginPro 2020 software was used for the PCA and HCA.

3. Results and Discussion

3.1. Species

Generally, we achieved the microgreens as good quality, ready to eat, but they differed in their growth dynamics. The fresh weight significantly depended on the species and the light treatment. It ranged from 8 mg (perilla) to 1052 mg (nasturtium) (Table S2, Figure 2A). The length of the plants (including the leaves) also depended on the species. The perilla plants were the shortest (2.0 cm), whereas the nasturtium plants were the longest (26.2 cm) (Table S2, Figure 3). Most of the plants were 5–8 cm in length. There were considerable differences in the biometric parameters of the plants on which the authors of earlier studies conducted their experiments, mostly because of differences in the time when the microgreens were harvested. Some researchers harvested the plants after they had produced cotyledons, but others harvested them only after the plants had produced the first true leaves, or, like in our research, after a certain number of days [18]. Due to the fact that the experiments were conducted on such small plants, the differences in the harvest time significantly influenced the research results. The seed sowing density may be



another factor determining the values of the parameters under analysis [19], likewise as light irradiance [20].

Figure 2. The fresh (**A**) and dry weight (**B**) [mg] for 28 microgreen crops depending on light combinations. The Red:Blue ratio for each combination–R light 6.7; B light 0.6; R + B light 1.6.



Figure 3. The length (including leaves) of the plants [cm] for 28 microgreen crops depending on light combinations. The Red:Blue ratio for each combination–R light 6.7; B light 0.6; R + B light 1.6.

The considerable differences in the values of the biometric parameters resulted mostly from the species characteristics of plants and their growth dynamics in the initial period of the growing season. The nasturtium, which forms very big and thick leaves, was characterised by the highest values of the biometric parameters and a high dry weight. On the other hand, the species which formed delicate leaves in the initial period of their growth, such as dill, coriander, Welsh onion, watercress, and rocket, gave much lower yields of fresh and dry matter. Interestingly, basil, especially lemon basil and 'Dark Opal', as well as perilla were characterised by low growth rates. This may have been caused by the fact that the growth temperature was too low for these thermophilic species. Another research also observed that sweet basil had a lower fresh weight than the other species under analysis [21].

The dry weight of the 28 microgreen crops ranged from 0.8 to 91.60 mg, as shown in Figure 2B and Table S2. The highest dry weight was noted for the nasturtium, and the lowest for the watercress. Most of the plants obtained 1.5–6.0 mg dry weight. There were similar results from the research conducted by Xiao [1]. These plants typically have a high water content [21]. The dry matter content increased as the plants matured [22].

3.2. Light Treatment

The light treatments affected the yield of the fresh and dry weight of microgreens in our experiment (Figure 2, Table S2). The biomass production did not increase in most of the species as a result of the B light treatment, but it did when the plants had been treated with R light. Red light significantly influenced the fresh mass in 15 species. The greatest differences (exceeding even five times) between the R light and the other combinations were observed in the purple kohlrabi, less also in mizuna, leaf chicory, and coriander. These results are in line with the findings of earlier studies [22,23]. However, the results of a significant number of the species (24) grown under the R + B light were similar to or worse than the results of those grown under the B light. Only the pea microgreens had a significantly higher fresh mass under the R + B light than the other combinations (the nasturtium also had a higher fresh mass, but the difference was not statistically significant). Red light is very efficient in the production of tall strong fast-growing plants. However, this effect can be observed when in the spectrum there is not much blue light, which obscures the chloroplast to protect it from too intense light [24]. This situation was observed in the R + B light combination, where red light dominated the spectrum, but blue light also had a high share. The exposure of dill, garden cress, garnet amaranth, green sweet basil, and perilla to the B light resulted in the highest fresh mass of these plants. And these results were really significantly higher than the other combinations. The authors of other studies conducted on various species of microgreens also observed the highest yield of fresh matter under blue light [25,26]. According to Macedo et al. [27], higher proportions of blue light are associated with the development of "sun type" leaves. This type of leaves results in an increase in leaf-blade area, the number of leaves, and leaf thickness, mainly due to an increase of the palisade parenchyma. The larger leaf intercepts more light, which may lead to a significant increase of biomass under low light intensity. Probably for this species the PPFD was too low. On the contrary, Pennisi et al. [12] conducted an experiment on basil and found that the spectrum with a high share of red light resulted in the highest yield of fresh matter contrary to another research [28]. However, plant yield can reach a plateau or decrease when the blue light proportion in the spectrum reaches a threshold, which varies among species [29]. In conclusion, the response to blue-red lighting is species-dependent, which has also been shown in other studies [28,30,31].

It is noteworthy that in our research there was about 10% green light in all light combinations. Kamal et al. [22] conducted a study on *Brassicaceae* microgreens and found that the plants exposed to supplemental illumination with green LEDs (R70: G10: B20) were characterised by better vegetative growth and morphology than the microgreens grown under red and blue light only. This may be due to the fact that phytochromes and cryptochromes are also green-light receptors [32].

The light treatment significantly influenced the dry weight. The spectra with high and very high shares of red light (R and R + B light) significantly increased the dry weight in the herb of 18 microgreen crops. Only three species had the highest dry weight under the B light.

The light diversified the length of the microgreens to a lesser extent (Figure 3, Table S2). The light treatment had no effect on the length of nine plant species. The spectrum with

the dominant share of red light significantly influenced the length of seven species of microgreens. The B light increased the length of only two species. The length of the plants grown under the R + B light was similar to those grown under the R light as well as those grown under the B light, except for the amaranth, where the tallest plants grew under the R + B light. The authors of other studies observed that the length of the hypocotyl depended not only on the R:B ratio but also on the species. The length of the hypocotyl in the plant species of the *Brassicaceae* family decreased along with the decreasing red/blue ratio [23]. Other authors also observed that a higher share of blue light in the blue and red spectrum significantly inhibited the elongation of the hypocotyl in microgreens [25]. Probably, the species difference in elongation promoted by R or B light is due to inherent genetic differences in response to shade [33].

3.3. H/DW Ratio

The calculation of the plant height to dry weight ratio H/DW (cm·mg⁻¹) revealed interesting results concerning the morphology of the plants (Figure 4). The highest value of H/DW was obtained by dill under R light (13.70), and the lowest by borage under B light (0.19). Most species ranged from 1 to 3. The R light resulted in the highest value of the H/DW ratio in 11 of the species. Only four microgreen crops had the highest value of the H/DW ratio under the B light and five under the R + B light. Also, for five species there were no differences in H/DW value for B and R + B light. The results of the research conducted by Toscano et al. [26] showed that the same amount of dry matter was distributed over longer plants grown under red light. This means that under these experimental conditions the hypocotyls were thinner, which affected the total plant biomass.



Figure 4. The H/DW ratio of 28 microgreen crops depending on light combinations. The Red: Blue ratio for each combination–R light 6.7; B light 0.6; R + B light 1.6.

3.4. PCA and HCA

The PCA results revealed a negative correlation between the type of spectral composition of light (light) and the length of the microgreens (length), and the H: DW ratio of the studied microgreen species (Figure 5). The positive correlation was between the examined



biometric parameters (length, fresh mass, and dry weight). The PCA analysis showed that the effect of light depended on the plant species.

Figure 5. Principal components analysis (PCA) of microgreens from biometrical analyses. Twodimensional (2D) scores plot of species of microgreens from different spectral compositions of light (red points) presented relationships between species and the measured biometric parameters.

The HCA showed similarities and differences between the microgreen species and the spectral composition of light within the parameters under analysis (Figure 6). Two groups of plants (purple and green lines) differing in the values of the biometric parameters were distinguished in the analysis. Another two groups with the greatest similarity (90–95%) were distinguished within the aforementioned two groups. The analysis showed that the spectral composition of light had a significant influence on the microgreen crops. The R light had the greatest effect on the radish, kale, spinach, and to a lesser extent, coriander and rocket. The B light–on the Indian mustard, garden cress, dill, and garnet amaranth, whereas the R + B light mixture–on the borage, green sweet basil, lucerne, and red cabbage. The location of microgreen from one spectral composition in another group (cluster) might indicate the preferences of this species for a specific spectral composition of light. The nasturtium, which formed a separate group (the light green line), was significantly different from the other microgreens. Moreover, a significant influence of the investigated spectral composition of light on the biometric parameters of the purple kohlrabi was found. Other researchers also observed species-specific responses to the quality of light [23,34].



Figure 6. Hierarchical clustering demonstrates biometric parameters of microgreens in relation to the spectral composition of light.

4. Conclusions

The results of our study showed that the light quality modulated different aspects of the growth of the microgreen crops. The biometric parameters of the plants were influenced by the spectra with an increased share of both red and blue lights. The species under analysis differed considerably in their responses to the applied light spectra. The spectrum with the highest share of red light resulted in higher fresh and dry weights of most of the species under study. The light treatments had a lesser influence on the length of the plants. The combinations with a higher share of red light also resulted in higher H/DW values. The R + B light combination with about 50% red light in the spectrum had the smallest influence on plant growth. The values of the biometric parameters of the nasturtium were significantly greater than those noted in the other microgreen species.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agriculture12050576/s1, Table S1: Twenty-eight microgreens assayed in the study; Table S2: Morphological parameters of individual species, Figure S1: The growth chamber.

Author Contributions: Conceptualization, B.F.; methodology, B.F.; software, M.K.-M.; writing original draft preparation, B.F. and M.K.-M.; writing—review and editing, B.F.; visualization, M.K.-M.; project administration, B.F.; funding acquisition, B.F. All authors have read and agreed to the published version of the manuscript. **Funding:** Publication was co-financed within the framework of the Polish Ministry of Science and Higher Education's program: "Regional Initiative Excellence" in the years 2019–2022 (No. 005/RID/2018/19)", financing amount 1,200,000 PLN.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: We are grateful to "W.Legutko" company (Poland) for providing seeds for the experiment.

Conflicts of Interest: The authors declare that there is no conflict of interest related to this article.

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