# Journal of Agricultural Sciences Research

## PRODUCTION OF LETTUCE, BEET AND MUSTARD MICROGREENS UNDER DIFFERENT KINDS OF LIGHT

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Abstract: Microgreens can be considered a class of very young plant food, with a delicate texture, rich in phytochemicals and which has a short stem with one or two pairs of rudimentary leaves. The objective of this work was to evaluate different species of vegetables and kinds of light in the production of microgreens. The seeds of different vegetable species were sown in plastic trays containing Carolina Soil® substrate. The treatments consisted of different species (lettuce, beet root and mustard) and kinds of light (red LEDs, blue LEDs, green LEDs and control). The experimental design used was a 2 x 4 factorial design, with five replications. After 18 days of germination, plant height, number of leaves, fresh and dry mass of the aerial part were evaluated. The data obtained were subjected to analysis of variance, with the means compared using the Tukey test at a 5% probability of error. The height of lettuce microgreens was greater than that of beet root, but it did not differ from mustard microgreens. The microgreens under the green LEDs (9.21 cm) were taller than those grown under the blue LEDs (8.57 cm), but did not differ between the other treatments. The highest number of leaves (4.28) of mustard microgreens was obtained under the red LEDs, but in lettuce and beet root there was no difference between treatments. The amount of fresh mass of mustard microgreens was greater than those obtained with lettuce and beet root in the different kinds of light, with the averages observed in the red LEDs being higher than those in the control treatment for the three species. The variable amount of Dry mass did not have a significant effect on the factors evaluated. The type of light and plant species influence the production of microgreens.

**Keywords:** Leds, Quality of light, Nutraceutical. Functional foods

## **INTRODUCTION**

Food production is essential for the survival of humanity and maintaining quality of life. Projection studies involving population growth indicate that the world population is expected to reach approximately 9.7 billion in 2050 (UN, 2024). It is known that as the global population grows, there is a greater need for food.

Currently, the increase in the world's population continues to be accompanied by an increase in agricultural production of commodity foods and supply will continue for many years to meet growing demand. However, it is worth highlighting that global agriculture already faces several challenges, including extreme climate and weather changes causing flooding and severe droughts in food production fields. As well as soil degradation, expansion of arid lands and increase in the price of inputs and transportation costs (BENKE & TOMKINS, 2017). Furthermore, projections indicate that in 2050 the arable area per capita will correspond to one third when compared to data from 1970 (FAO, 2024).

New technologies are revolutionizing agriculture, with an innovative and sustainable approach, optimizing space and productivity, reducing dependence on external factors. As an example, we have the technological advances in the manufacturing materials of LEDs (Light Emitting Diode) at the end of the 90s, resulting in a new generation with high brightness. This way, they began to be used as a source of lighting. Until then, they were only used as an on/off indicator in electronic equipment due to the low amount of light emitted (ZHELUDEV, 2007).

This change in the amount of light emitted by LEDs has generated new interests and commercial applications, such as use in car headlights and ambient lighting. In plant production, they were initially used in in vitro production growth rooms (OLIVEIRA, et al., 2011). And later, in the in-door production of hydroponic vegetables, baby leaves and microgreens grown in closed environments without receiving natural light, such as buildings located on the outskirts of large urban centers (GEROVAC, et al., 2016).

It is known that light is one of the most important environmental factors in vegetable cultivation, as it has a strong influence on plant growth and development. Therefore, the quality of light, that is, wavelength, light intensity and photoperiod has a great impact on the morphology and metabolism of the plant (YANG et al., 2018).

In recent years, the demand for fresh and functional foods has grown. In 2021, microgreens sales worldwide were estimated at US\$1.8 billion and by 2028 they are expected to exceed US\$2 billion (ALLIED MARKET RESEARCH, 2024). In more sophisticated markets, microgreens have gained popularity (ABAAJEH et al., 2023). This increase in the consumption of microgreens has been driven by the growing interest of consumers in incorporating foods into their diet that can meet nutritional needs and also contribute to health and longevity (ZHANG et al., 2020; KYRIACOU et al., 2016).

Studies show that diets rich in vegetables and fruits are associated with lower rates of cancer, chronic diseases such as cardiovascular diseases, stroke, diabetes and hypertension. This is due to the fact that these kinds of foods contain compounds with antioxidant properties, such as flavonoids, glucosinates, isoflavones and terpenes (DREWNOWSKI & GOMES-CARNEROS, 2000; EBERT, 2022). The Word Health Organization (WHO) recommends a daily intake of 400 g of fruits and vegetables to prevent chronic diseases and micronutrient deficiencies (WHO, 2024).

Within the list of functional foods that can promote the benefits mentioned above, microgreens can be highlighted, which are obtained from these edsof different plantspecies (HADDAJI, et al., 2023), such as the Brassica vegetable oleracea (ROCHA et al., 2024). Microgreens can be understood as young, tender plants with the first pair of definitive leaves still rudimentary, produced from seeds without treatment with agrochemicals. Microgreens stand out compared to adult plants because they can contain amounts of phytochemicals considered beneficial to health in concentrations that can exceed up to 40x (SAMUOLIENĖ, et al., 2013).

Microgreens are among the kinds of cultivated plants that can be produced efficiently in environments such as biofactories or greenhouses with controlled light, temperature and humidity.

These cultivation facilities can be located in urban and peri-urban environments, since microgreens are compact plants, allow vertical cultivation, have a short cycle (harvest 15 to 21 days after germination), require reduced amounts of resources for cultivation maintenance and have a high market value (KOZAI & NIU, 2020). Given the above, the objective of this work was to evaluate different species of vegetables and kinds of light in the production of microgreens.

### **MATERIAL AND METHODS**

The present work was conducted in a growth chamber of the Department of Agricultural Sciences at ``Universidade Regional Integrada do Alto Uruguai e das Missões`` – URI Erechim.

To conduct the experiment, transparent trays made of polystyrene with dimensions of 11 x 11 x 3 cm were used, containing 37 g of commercial Carolina Soil® substrate. In each tray containing the previously moistened substrate, 1.0 g of seed was sown, uniformly, over the surface. The trays containing the seeds were kept inside the growth chamber with a roof that allows sunlight to pass through, with

a temperature of 25°C + 1°C. After emergence, the trays were placed on benches 60 cm from the light source with a 16-hour photoperiod. Throughout the cultivation period, watering was carried out daily with the aid of a pick, in order to meet the plants' water needs.

The treatments used consisted of different kinds of light [red LEDs EDER 3LA3 630nm, blue LEDs EDEB-3LA1 470nm, green LEDs 3LA1 530nm and control treatment (absence of artificial light)] and vegetable species (lettuce, beet root and mustard). The experimental design used was a 3 x 4 factorial design (Kinds of light x Species), with six replications per treatment, with the experimental unit consisting of a tray. After 20 days of germination, the height of the plants, the number of leaves per plant, the amount of dry mass and dry mass of the microgreens were evaluated.

The height of the plants was measured using a graduated ruler. To determine the amount of fresh mass of the microgreens, a cut was made in the stem of the plant, with the help of scissors, approximately 1 cm from the surface of the tray's cultivation substrate.

The cut plant material was weighed on a digital scale and then placed in a previously identified paper bag. Subsequently, the bags containing the plant material were placed in a forced aeration oven at a temperature of 60 °C for 72 hours.

The data obtained were subjected to analysis of variance, with the means compared using the Tukey test at a level of 5% probability of error using the Saneste statistical program (ZONTA & MACHADO, 1992). Data relating to the number of leaves were transformed into  $(x + 0.5) \frac{1}{2}$ , and data on shoot length, fresh and dry weight were not transformed.

### **RESULTS AND DISCUSSION**

According to the analysis of variance, there was a significant effect of the factors evaluated, species and type of light, for the variable average height of the microgreen's plants. However, there was no interaction between the factors. It was observed (Table 1) that the average height of lettuce plants (9.1 cm) was higher than the averages obtained for beet plants (8.48 cm) (Table 1). However, when comparing the height of beet root and mustard plants, no difference was observed between them.

Species	Plant height (cm)
Lettuce (Lactuca sativa L.)	9,10 a
Beet root (Beta vulgaris L.)	8,48 b
Mustard [Brassica juncea (L.) Czern]	8,84 ab
CV (%)	6,27

Table 1: Height of microgreens plants of different plant species, after 20 days of germination.

\* Means followed by the same letter in the column do not differ significantly from each other at the 5% error probability level using the Tukey test.

In general, the average plant heights (8.48 to 9.10 cm) obtained in the production of microgreens with different species of vegetables can be considered adequate. The difference observed in the average height of the plants between the three species can be attributed to the genetic factor. According to Bulgari et al. (2017), plant height can vary from 2.5 to 8 cm. These values may vary according to the species, type of substrate, growing conditions and cycle, which can vary from 7 to 21 days after seed germination.

Regarding the effect of the type of light on plant height, it can be observed that the microgreens grown under the green LEDs (9.21 cm) presented higher averages than those obtained with the microgreens grown under the blue LEDs (8.57 cm); (Table 2). These results differ from those obtained

by Grishchenko et al. (2022), who, when comparing micro propagated potato plants under red, blue and green LEDs, observed that plants grown under red LEDs had a greater height (150 mm) than those grown under green LEDs and blue LEDs. (106 mm and 67 mm, respectively).

Kinds of light	Plant height (cm)
Red LEDs EDER 3LA3 630nm	8,78 ab
Blue LEDs EDEB-3LA1 470nm	8,57 b
Green LEDs 3LA1 530nm	9,21 a
Control treatment	8,63 ab
CV (%)	6,27

Table 2: Height of microgreens plants grown under different kinds of light after 20 hours of germination.

Still in Table 2, it is noted that there was no difference between the means obtained with microgreens grown under red LEDs (8.78 cm), blue LEDs (8.57 cm) and control treatment (8.63 cm). Different results were obtained by Rocha et al. (2013) who, working on the in vitro multiplication of blackberry shoots under different kinds of light, found that the height of the shoot did not differ between blue, red and green LEDs.

On the other hand, Brazaitytė et al. (2021) observed that blue LEDs negatively interfere with the height of microgreens. The aforementioned authors, evaluating the effect of different percentages of blue LEDs and red LEDs on the production of mustard (*Brassica juncea* 'Red Lace') and kale (*Brassica napus* 'Red Russian') microgreens, observed that the increase in the percentage of blue LEDs in the environment of cultivation contributed to the reduction of elongation of both species.

The type of light considered most suitable may vary depending on the species to be used in the production of microgreens. Well, Toscano et al. (2021), evaluating the effect of different light spectrums on the production of microgreens of two species, obtained interaction between the factors. Therefore, the height of turnip plants (*Brassica rapa L. subsp. oleifera*) grown under blue LEDs was greater than those grown under red LEDs. On the other hand, there was no difference in the height of Amaranth microgreens (*Amaranthus tricolor L.*) when comparing the type of light.

Regarding the variable number of leaves per plant, there was an interaction between the factor's species and type of light. It was observed that the highest average was obtained in mustard microgreens (2.95 leaves per plant) occurred under red LEDs (Table 3), and there was no difference between the other treatments. This result was not repeated in lettuce and beet root microgreens, as the number of leaves per plant did not differ between different kinds of light. The results observed in this work suggest that the type of light used in the growing environment may increase the number of leaves formed per microgreen plant. Furthermore, it shows that the influence of light on the formation of new leaves in the plant depends on different spectra (450 to 750 nm) and also on the plant species. Therefore, the lighting condition considered optimal for one species will most likely present different results with another species, varying according to the plant species used in production.

<sup>\*</sup> Means followed by the same letter in the column do not differ significantly from each other at the 5% error probability level using the Tukey test.

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Kinds of light	Lettuce	Beet root	Mustard
Red LEDs EDER 3LA3 630nm	4,28 aA	2,02 aC	2,95 aB
Blue LEDs EDEB- 3LA1 470nm	4,41 aA	2,17 aB	2,09 bB
Green LEDs 3LA1 530nm	4,23 aA	2,00 aB	2,00 bB
Control treatment	4,65 aA	2,10 aB	2,06 bB
CV (%)			3,36

Table 3: Average number of leaves formed by lettuce, beet root and mustard microgreens plants grown under different kinds of light after 20 days of germination.

Stefanel et al. (2020), evaluating the effect of different kinds of light (blue LEDs, green LEDs, red LEDs, white LEDs and fluorescent lamps) on the multiplication of wild cherry (*Eugernia involucrata*) shoots, they found that the largest number of leaves per shoot occurred in those grown under blue LEDs and fluorescent lamps (4.00 and 3.89, respectively). Different results were obtained by Asdame et al. (2022), who observed after 24 days of sowing that lettuce plants cultivated Levistro and Carmoli cultivated under blue LEDs had a greater number of leaves (6.9 leaves per plant) than those cultivated under red LEDs (5.7 leaves per plant).

As for the variable quantity of fresh mass, according to analysis of variance there was an interaction between the factors (species x type of light). The amount of fresh mass obtained with mustard microgrees was higher than that observed in lettuce and beet root under different kinds of light (Table 4). Still in the same Table, it can be noted that there was no difference in the amount of fresh dough between the kinds of LEDs used in the production of beet root and mustard microgreens. On the other hand, in lettuce microgreens the amount of fresh

mass obtained under the red LEDs (22.45 g) was greater than that coming from the green LEDs (17.68 g), but there was no difference between the averages obtained between LEDs blue (20.81g) and red LEDs.

Distinct results were obtained Toscano et al. (2021), which evaluated the production of microgreens from amaranth (*Amaranthus tricolor L.*) and green turnip [(*Brassica rapa L*). subsp. Oleifera], under different kinds of LEDs (white, blue and red), obtained a significant effect of the factors, but there was no interaction between them. Furthermore, they observed that the greatest amount of fresh mass occurred with microgreens grown under blue LEDs.

Kinds of light	Lettuce	Beet root	Mustard
Red LEDs EDER 3LA3 630nm	22,85 aB	13,57 aC	48,00 aA
Blue LEDs EDEB- 3LA1 470nm	20,81abB	11,52 aC	48,12 aA
Green LEDs 3LA1 530nm	17,68 bB	11,42 aC	49,86 aA
Control treatment	9,85 cB	10,62 aB	41,63 bA
CV (%)			9,01

Table 4: Quantity of fresh mass of lettuce, beet root and mustard microgreens, from cultivation under different kinds of light, after 20 days of germination.

Putri et al. (2023) evaluating the production of microgreens from three species [amarantus (Amaranthus cruentus L.), coriander (Coriander sativum L.) and red radish (Raphanus sativum L.) under different kinds of light (white, blue, purple and red), obtained a significant effect of the factors and realized that the greatest amount of fresh mass was obtained with radish microgreens. Regarding the type of light, there was no difference between them in horseradish and coriander

<sup>\*</sup> Means followed by the same letter, lowercase in the column and uppercase in the row, do not differ significantly from each other, at the level of 5% probability of error using the Tukey test.

<sup>\*</sup> Means followed by the same letter, lowercase in the column and uppercase in the row, do not differ significantly from each other, at the 5% error probability level using the Tukey test.

microgreens. On the other hand, he noted that in amarantus the highest averages were achieved with red light and purple light.

Still in Table 4, it can be seen that the production of fresh mass of lettuce microgreens was stimulated through the use of LEDs. The averages obtained under the red LEDs (22.85 g) were higher than those under the green LEDs (9.85 g) and control treatment (9.85 g).

Additionally, although there was no statistical difference between the different kinds of LEDs in beet root microgreens, it can be seen that the amount of mass in all kinds of light was higher than the control treatment (absence of artificial light). According to Dorais & Gosselin (2002), the production of microgreens in protected environments can be limited by the quality of natural light (photoperiod and light intensity). Therefore, for the production of lettuce microgreens in a protected environment with natural light, it may be necessary to supplement artificial light to increase the photoperiod.

Similar results were obtained by Sumi et al. (2024) who, evaluating the production of lettuce microgreens under different lighting conditions with LEDs, observed that the fresh mass of 100 plants produced under red and blue LEDs did not differ statistically. Furthermore, they also found that microgreens production may vary in the amount of fresh mass within the same species (Lactuca sativa L.), as red lettuce presented a greater quantity than green lettuce (4.36 and 4.23 g per 100 plants).

Regarding the amount of dry mass, there was no significant effect for the factors evaluated. It can be seen in Table 5 that the amount of Dry mass did not differ statistically between the different kinds of light. However, from the point of view of absolute value, the highest average was obtained from microgreens grown under the red LEDs (2.31 g) and the smallest in the control treatment (1.14 g). Similar results were obtained by

Brazaitytė et al. (2021), with the production of mustard (Brassica juncea 'Red Lace') and kale (*Brassica napus 'Red Russian*') microgreens under different kinds of light, which observed that there was no statistical difference in the amount of Dry mass produced with the microgreens grown under the red LED's and blue LED's.

Kinds of light	Dry mass (g)
Red LEDs EDER 3LA3 630nm	2,31 a
Blue LEDs EDEB-3LA1 470nm	1,48 a
Green LEDs 3LA1 530nm	1,43 a
Control treatment	1,14 a
CV (%)	80,7

Table 5: Quantity of dry mass of microgreens from cultivation under different kinds of light, after 20 days of germination.

\* Means followed by the same letter in the column do not differ significantly from each other at the 5% error probability level using the Tukey test.

In Table 6, it is noted that the amount of dry mass did not differ between the species evaluated. Different results were obtained by Toscano et al. (2021), who, when evaluating dry mass production, obtained a difference between species, with the highest average obtained with turnip (*Brassica rapa L. subsp. oleifera* (DC.) Metzg) and the lowest with amaranth (*Amaranthus tricolor L.*). Possibly, the responses promoted by LEDs for this variable may vary depending on the plant species used in the production of microgreens.

Species	Dry mass (g)
Mustard [Brassica juncea (L.) Czern]	2,08 a
Lettuce (Lactuca sativa L.)	1,59 a
Beet root (Beta vulgaris L.)	1,10 a
CV (%)	80,7

Table 6: Quantity of dry mass of microgreens from the cultivation of different species, after 20 days of germination.

<sup>\*</sup> Means followed by the same letter in the column do not differ significantly from each other at the 5% error probability level using the Tukey test.

### **CONCLUSIONS**

For the conditions under which the experiment was conducted, it can be concluded that:

- the height of the plant and the amount of fresh mass of microgreens may vary according to the plant species and the type of light used in the growing environment.

- the type of light and plant species influences the production of microgreens.

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