**Revista em Agronegócio e Meio Ambiente – RAMA**
DOI: 10.17765/2176-9168.2024v17n.Especial.e12444
e-ISSN 2176-9168
*Tecnologias Limpas*

# Colored led reduces energy use, affecting lettuce seed germination, growth, and antioxidant activity positively

## *LED coloridas economizando energia, afetando positivamente a germinação, crescimento e atividade antioxidantes em alface*

**Ana Luiza Reale[1], Heloisa Santos[2], Giovani Tirelli[3], Leticia Moreno[4], Wilson Pereira[5], Elisa Bicalho[6]**

**ABSTRACT**: As vegetables have been growing and space has gathered on the market, there is increasing demand for alternative light sources. This research aimed to evaluate the effects of colored LEDs on the germination and initial growth of lettuce plants, as well as their effects on the antioxidant system. Seeds were germinated in a chamber at 20°C under a 12-hour photoperiod. The light treatment in the first phase consisted of white and colored light-emitting diode (LED) lights (red-V + blue-A) in proportions of 100% V, 80% V + 20% A, 50% V + 50% A, and 80% A + 20% V. The first phase of the experiment consisted of a completely randomized design in a 2x4 factorial scheme (two light conditions and four seed lots) with four replications. The first count, germination, germination speed index (GSI), root length, shoot length, total seedling length, and shoot-to-root ratio were evaluated via image analysis. For the second phase of the experiment, the quantification of antioxidative enzyme activity (SOD, CAT, and APX) was performed to assess whether the light treatments (white LED light, colored LED light, and fluorescent light) caused photooxidative damage in the seedlings. Compared with white LED light, colored LED light improved plant germination and growth by promoting faster radicle protrusion, a greater GSI, a longer total seedling length, and a longer primary root length. The quantification of SOD, CAT, and APX activity indicated that the quality of light used in this work did not cause photooxidative stress in lettuce plants.

**Keywords:** Antioxidant system; *Lactuca sativa*; Light emitting diode; Photooxidation; Seed quality.

**RESUMO:** À medida que as hortaliças vêm crescendo e ganhando espaço no mercado, há uma demanda crescente por fontes alternativas de luz. Esta pesquisa teve como objetivo avaliar os efeitos dos LEDs coloridos na germinação e no crescimento inicial de plantas de alface, bem como seus efeitos no sistema antioxidante. As sementes foram germinadas em câmara a 20°C sob fotoperíodo de 12 horas. O tratamento luminoso na primeira fase consistiu em luzes de diodo emissor de luz (LED) brancas e coloridas (vermelho-V + azul-A) nas proporções de 100% V, 80% V + 20% A, 50% V + 50% A e 80% A + 20% V. A primeira fase do experimento consistiu em delineamento inteiramente casualizado em esquema fatorial 2x4 (duas condições de luz e quatro lotes de sementes) com quatro repetições. A primeira contagem, germinação, índice de velocidade de germinação (IVG), comprimento da raiz, comprimento da parte aérea, comprimento total das plântulas e relação parte aérea/raiz foram avaliados por meio de análise de imagens. Para a segunda fase do experimento foi realizada a quantificação da atividade das enzimas antioxidantes (SOD, CAT e APX) para avaliar se os tratamentos luminosos (luz LED branca, luz LED colorida e luz fluorescente) causaram danos fotooxidativos nas mudas. Em comparação com a luz LED branca, a luz LED colorida melhorou a germinação e o crescimento das plantas, promovendo uma protrusão mais rápida da radícula, um maior GSI, um maior comprimento total de mudas e um maior comprimento de raiz primária. A quantificação da atividade de SOD, CAT e APX indicou que a qualidade da luz utilizada neste trabalho não causou estresse fotooxidativo em plantas de alface.

**Palavras-chave:** Diodo emissor de luz; Foto oxidação; *Lactuca sativa;* Qualidade de semente; Sistema antioxidante.

[1] Mestra em Agronomia (Fitotecnia) pela Universidade Federal de Lavras (UFLA).
[2] Doutora em Agronomia (Fitotecnia) pela Universidade Federal de Lavras (UFLA). Docente adjunta da Universidade Federal de Lavras (UFLA), Lavras, MG, Brasil.
[3] Mestrando em Agronomia pela Universidade Federal de Lavras (UFLA).
[4] Doutora em Agronomia pela Universidade da Georgia - Campus Tifton, GA, EUA.
[5] Doutor em Agronomia (Fitotecnia) pela Universidade Federal de Lavras (UFLA).
[6] Doutora em Biologia Vegetal pela Universidade Federal de Minas Gerais (UFMG). Docente permanente do Programa de Pós-Graduação em Fisiologia Vegetal da UFLA, Lavras, MG, Brasil.

**Autor correspondente:** Leticia Moreno
E-mail: leticia.moreno@ufla.br
*Recebido em: 02/02/2024*
*Aceito em: 21/05/2024*

## 1 INTRODUCTION

Currently, lettuce (*Lactuca sativa* L.) is the most consumed leaf vegetable worldwide, and in addition to its center of origin (the Mediterranean region), it must be cultivated outside of its center of origin to meet the increasing demand for consumption and, consequently, production (Sala; Costa, 2012). Therefore, plants will be exposed to unusual conditions from those that the plant is accustomed to developing (Sabir; Singh, 2013). Among all the environmental conditions, water and temperature are highly important for plant development (Taiz *et al.*, 2017). In addition, in lettuce, light sensitivity is crucial, with some studies showing that some physiological processes are stimulated by the presence of light (An; Zhou, 2017).

Lettuce seeds can be classified as photoblastic positive, negative, or neutral (not influenced by light) according to their response to the stimulus of light (Metivier; Viana, 1979). The influence of light on seed germination is directly correlated with phytochrome, which is a protein pigment photoreceptor of light (Yang; Pei, 1997). The effect of red light on the germination of seeds can be explained by the relationship between phytochromes and the synthesis of hormones (Peng *et al.*, 1997). Light activates phytochromes, initiating a signaling cascade that results in several physiological responses (Lovegrove; Hooley, 2000). In addition to red light, blue light is another color that plays an important role in plant phototropism, controlling the development of the plant hypocotyl, carotene synthesis, and stomatal opening. Studies of lettuce and alternating red and blue light during seedling development have shown that these light sources lead to an increase in plant and leaf growth as well as an increase in ascorbic acid and nitrate content, indicating improved nutritional value (Chen *et al.*, 2017).

Controlled environmental agriculture (CEA) is a method of crop production that is widely used in horticulture, where it is possible to control parameters such as temperature, humidity, water, and light exposure time (Liao *et al.*, 2020). An artificial light supply, common in CEA, has a low heat emission property and the possibility of light-spectrum control. Light-emitting diodes (LEDs), a type of artificial light, are currently being used to replace common light bulbs once LEDs significantly reduce energy consumption (Singh *et al.*, 2015). Just as excess natural light can cause reactions in seeds, the use of artificial lights in controlled environmental production will also affect seeds.

Furthermore, in agriculture, the use of LEDs can cause several reactions, resulting in accelerated plant growth. Despite being desirable, this growth acceleration results in the production of reactive oxygen species (ROS), and excess ROS lead to photooxidative stress (Morrow, 2008). One way to measure the damage caused by photooxidative stress is to observe the behavior of enzymes that are part of the plant antioxidant system. When the system is not able to eliminate or avoid ROS, the high accumulation of these free radicals may cause lipid peroxidation in cell membranes, such as damage to proteins, DNA, and other organic molecules, culminating in irreversible cell damage (Sevengor *et*

*al.*, 2011). The antioxidant system, through enzymes and secondary metabolites, acts by removing or limiting ROS formation to avoid oxidative stress (Sharma *et al.*, 2012). Superoxide dismutase (SOD), catalase (CAT), and ascorbate peroxidase (APX) are some examples of enzymes that are part of the antioxidant system (Havir and McHale, 1990).

Several studies have been performed to understand the effect of LEDs on the growth and productivity of different crops, such as wheat, lettuce, radish, and spinach (Yorio *et al.*, 2001). However, there is a lack of information on how LEDs can positively affect the initial stages of plant development and on the effect of LEDs on plant ROS production. With this in mind, the objective of this research was to evaluate the effects of colored LEDs on the germination and initial growth of lettuce plants, as well as their effects on the antioxidant system.

## 2 MATERIAL AND METHODS

The research was conducted at the Central Laboratory of Seed Research (LCPS on the Portuguese acronym) from the Federal University of Lavras (UFLA on the Portuguese acronym), with four samples (seed lots from 1 to 4) of crispy lettuce of Veronica's variety. The experiment was performed using two types of environmental lighting, white and colored, provided by chamber type B.O.D. (biochemical oxygen demand). The experiment was divided into two phases. In the first phase, physiological tests were performed on seeds from two types of light treatments. In the second phase, the best lot was chosen, and physiological and biochemical analyses were performed on seeds from the three types of light treatment.

### 2.1 FIRST PHASE

For the first phase, the seeds from each lot were disinfected in a sodium hypochlorite solution at a concentration of 0.02% for 30 seconds and subsequently washed in running water for 3 minutes. After that, the plants were allowed to germinate for seven days under two light conditions: white and colored. White light was provided by a B.O.D. instrument equipped with four white tubular LEDs, which included all wavelengths in the visible spectrum. For colored lights, the B.O.D. was equipped with four tubular LEDs in blue and red colors using the light color with a wavelength combination scheme according to Table 1. The light-changing scheme was determined according to the literature, which states that red light favors radicle protrusion (Sánchez *et al.*, 1990), while blue light stimulates elongation (Lin, 2002).

**Table 1.** The effect of light-changing scheme on the BOD in response to colored light treatment in the lettuce seed germination test

| Day | Light color | Wavelength |
|---|---|---|
| 1 | Red 100% | 625 nm |
| 2 | Red 100% | 625 nm |
| 3 | Red 80% + Blue 20% | 580 nm |
| 4 | Red 50% + Blue 50% | 510 nm |
| 5 | Red 50% + Blue 50% | 510 nm |
| 6 | Red 20% + Blue 80% | 460 nm |
| 7 | Blue 100% | 440 nm |

As part of the first phase, the following analyses were performed:

a. Germination test

The test was performed by placing seeds on germination paper moistened with distilled water (at a rate of 2.5 times the paperweight) inside an acrylic container (Brasil, 2009). The analysis was performed using four repetitions with 50 seeds each. The B.O.D. chamber was set at a constant temperature (20°C) and a photoperiod of 12 hours (for both white and colored light environments). Germination seed evaluation was performed on the fourth (first count) and seventh days (final count), and the results are expressed as the percentage of normal seedlings (all healthy structures of leaves, stems, and radicles) (Brasil, 2009). In addition to the germination test, the germination speed index (GSI) was also evaluated by counting the number of germinated plants daily. The index was calculated according to the method proposed by Maguire (1962).

b. Image analysis

Plant development was evaluated through images collected with GroundEye® (version S800) equipment; 10 normal seedlings from each replicate were collected on the fourth and seventh days during the germination test. Image analysis was used to measure the sizes of the primary roots and shoot and to determine the total length and shoot-to-root ratio of the plants.

## 2.2 SECOND PHASE

After the first phase, lot four was the higher-physiological quality lot selected for new physiological and biochemical tests. For this second phase, three light treatments were used: white tubular LED, fluorescent LED, and colored LED according to the light-changing scheme shown in Table 1. The seeds were allowed to germinate for seven days under three light conditions, and the germination test was performed using the protocol mentioned previously in the first phase according to Brasil (2009). In addition to the germination test, as part of the second phase, the following analyses were performed:

a. Biochemical analyses

The analyses were performed using seedlings from all 3 light treatments collected on the seventh day from the germination test of the second phase. Seeds from the higher-physiological quality lot were used as a control treatment for the biochemical analyses. Vegetative material from the seeds and seedlings was macerated using liquid nitrogen and polyvinylpyrrolidone (PVP) immediately after collection (7th day), and the macerated material was kept in a deep freezer (-86°C) until analysis was performed.

The antioxidant enzyme activities of SOD, CAT, and APX were quantified using the methodology proposed by Biemelt *et al.* (1998). A quantity of 200 mg of macerated sample was mixed with 1.5 mL of extraction buffer. The buffer was composed of 100 mM potassium phosphate (pH 7.8), 0.1 mM EDTA, and 10 mM ascorbic acid. The material was homogenized on a vortex and centrifuged at 12000 rpm for 30 minutes at 4°C, after which the supernatant was collected and used for determination of enzyme activity. The enzymatic activities of SOD, CAT, and APX were analyzed via electrophoresis.

SOD activity was measured by the ability of the enzyme to inhibit the photochemical reduction of nitroblue tetrazolium (NBT), as proposed by Giannopolits and Ries (1977). A volume of 10 µL of enzyme extract was combined with 190 µL of incubation media (composed of 50 mM potassium phosphate (pH 7.8), 14 mM methionine, 0.0001 mM EDTA, 0.075 mM NBT, and 0.002 mM riboflavin). The reaction media were kept in the dark until sample application. The tubes containing a mixture of buffer and sample, such as control tubes (incubation medium without the sample), were illuminated with a 20 W fluorescent lamp for 7 minutes at room temperature. After that, the absorbance was measured on a spectrophotometer at a wavelength of 560 nm. A unit of SOD activity was defined as the amount of enzyme that inhibited 50% of the NBT reduction ratio.

CAT activity was measured by the decrease in absorbance at 240 nm every 15 seconds for 3 minutes, which was monitored by hydrogen peroxide consumption (Havir and McHale, 1990). For that, 10 µL of the samples were mixed with 180 µL of incubation media (100 mM potassium phosphate, pH 7.0) previously heated at 30°C. A volume of 9 µL of 250 mM hydrogen peroxide (at a final concentration of 12.5 mM) was added to the mixture, which was immediately analyzed on a spectrophotometer at a wavelength of 240 nm. A unit of CAT activity was defined as the amount of enzyme necessary to decompose 1 µmol per minute of H2O2¬.

APX was measured by the decrease in ascorbate absorbance (ε = 2,8 mM-1 cm-1) in a spectrophotometer at 290 nM every 15 seconds for 3 minutes. A sample of 9 µL was added to 180 µL of preheated (30°C) reaction media composed of 100 mM potassium phosphate (pH 7.0) and 0.5 mM ascorbic acid. After mixing the sample with the incubation media, a volume of 9 µL of 2 mM hydrogen peroxide (final concentration of 0.1 mM) was immediately read on a spectrophotometer (Nakano; Asada, 1981). APX activity was defined by the amount of enzyme required to oxidize 1 µmol per minute of ascorbic acid.

### 2.3 STATISTICAL ANALYZES

The experimental design was a completely randomized design (CRD), with a factorial scheme of 2 (treatments) × 4 (seed lots) for the first phase, and a CRD with a factorial scheme of 3 (treatments) × 1 (seed lot) for the second phase. All the data were analyzed by the software SISVAR® (Ferreira, 2019) through analysis of variance (ANOVA), and the means were compared using Tukey's test at 5% probability.

## 3 RESULTS AND DISCUSSION

The results from the first germination count showed no significant differences between light treatments for lots 1 and 2; however, for lots 3 and 4, greater germination at the first count was observed under colored lights (Figure 1a). However, the total percentage of germination (final count) did not significantly differ when the light conditions were analyzed (Figure 1b). For the GSI, in addition to that in lot 4, all the lots exhibited significant differences among the light treatments, where a greater index was observed for lots under colored light (Figure 1c).

Seed vigor can be measured in different ways, such as by the germination speed index and the first germination count, where taller vigor seeds germinate faster than lower vigor seeds (Franzin *et al.*, 2004). The results presented here indicate that red light during the first few days after sowing promotes faster radicle protrusion than white light. This can probably be explained by the direct action of red light on the gibberellin-abscisic acid balance (Olszewski; Sun; Gubler, 2002).

While gibberellin (GA) promotes germination and consequently overcomes quiescence and dormancy, abscisic acid (ABA) has the opposite effect (Finch-Savage and Leubner-Metzger, 2006). Thus, the results observed here may be linked to the fact that light quality acts directly on the GA-ABA balance, which is part of the photorefractive process. The photoreceptor phytochrome B (phyB) is active (Pfr) when receiving red light or inactive (Pr) when the stimulus comes from distant red light (Seo *et al.*, 2006). In Arabidopsis seeds, Pr, which is inactive due to the accumulation of distant red light, modulates the balance of GA/ABA by stimulating the transcription of DELLA proteins, a group characterized as negative GA regulators (Tyler *et al.*, 2004).

As shown in Figure 1a, on the first count, higher values were found for lots 3 and 4 under colored lights than under white lights. This finding indicates a possible connection between colored light and fast radicle protrusion only on high-quality seed lots, as colored light does not influence low-quality seed lots (1 and 2). Therefore, although colored light stimulates the speed of seed germination, it does not promote an increase in physiological seed quality because the final percentage of germination was not influenced by light treatment (Figure 1). Similar results were found by Solano *et al.* (2021) for melon and pea seeds, where red light did not increase the percentage of germination but was effective at promoting precocious germination (faster germination).

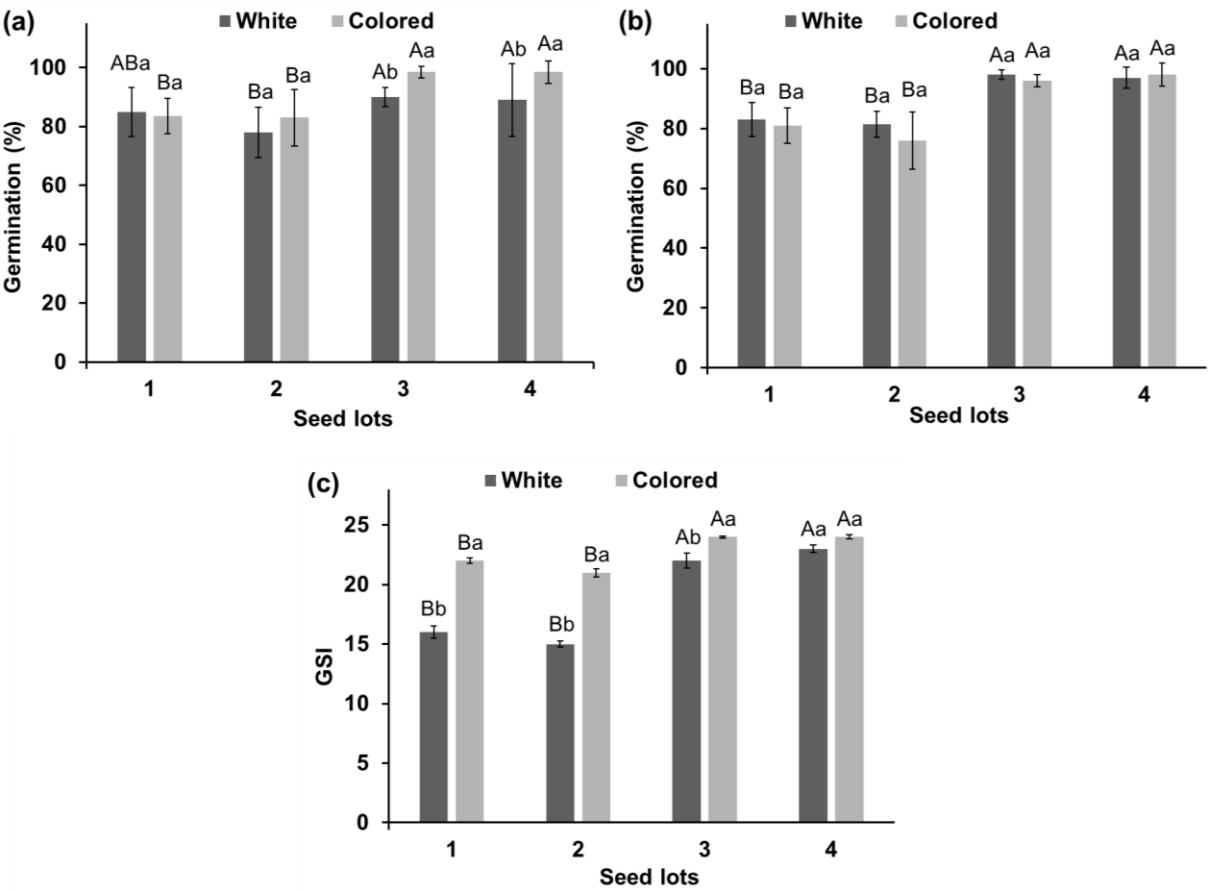


**Figure 1.** Percentage of first count (a), final count (b), and GSI (c) of *Lactuca sativa* under two light treatments.

The means followed by equal letters, uppercase letters comparing seed lots inside each light spectrum and lowercase letters comparing each lot between the two light spectra, are not significantly different according to Tukey's test ($p \leq 0.05$)

The results of seedling growth obtained by using the GroundEye® equipment can be observed in Figure 2. The shoot length was not influenced by light treatment, except for lot 1, where white light resulted in greater shoot values than did the colored light treatment (Figure 2a). A positive effect of white light on shoot length was reported by Shimizu *et al.* (2011), who reported that, compared with mixed lights, monochromatic lights increased shoot length. Additionally, Ohashi-Kaneko *et al.* (2007) reported greater hypocotyl growth in lettuce plants under white light. Overall, the results presented here may indicate that although colored lights can increase lettuce seed germination, it is important to use monochromatic light after germination to increase shoot growth and consequently yield.

The root length, seedling length and shoot-to-root ratio were positively influenced by the colored light treatment, with all the lots exhibiting greater values under colored light than under white light (Figures 2b, 2c, and 2d). An increase in seedling length due to different light conditions has already been reported for other species, such as Mentha piperita, M. spicata, M. longifolia (Sabzalian *et al.*, 2014), and Brassica oleracea (Paniagua-Pardo *et al.*, 2015). Sabzalian *et al.* (2014), studying Mentha sp., showed that the

combination of 70-30% red–blue light resulted in an increase in fresh weight, the photosynthesis ratio, and essential oil yield compared to those under field conditions. By analyzing lettuce plants under artificial light, Lin *et al.* (2013) reported that a combination of white and colored light (red and blue) resulted in greater weight (fresh and dry), better plant morphology and better flavor than light alone.

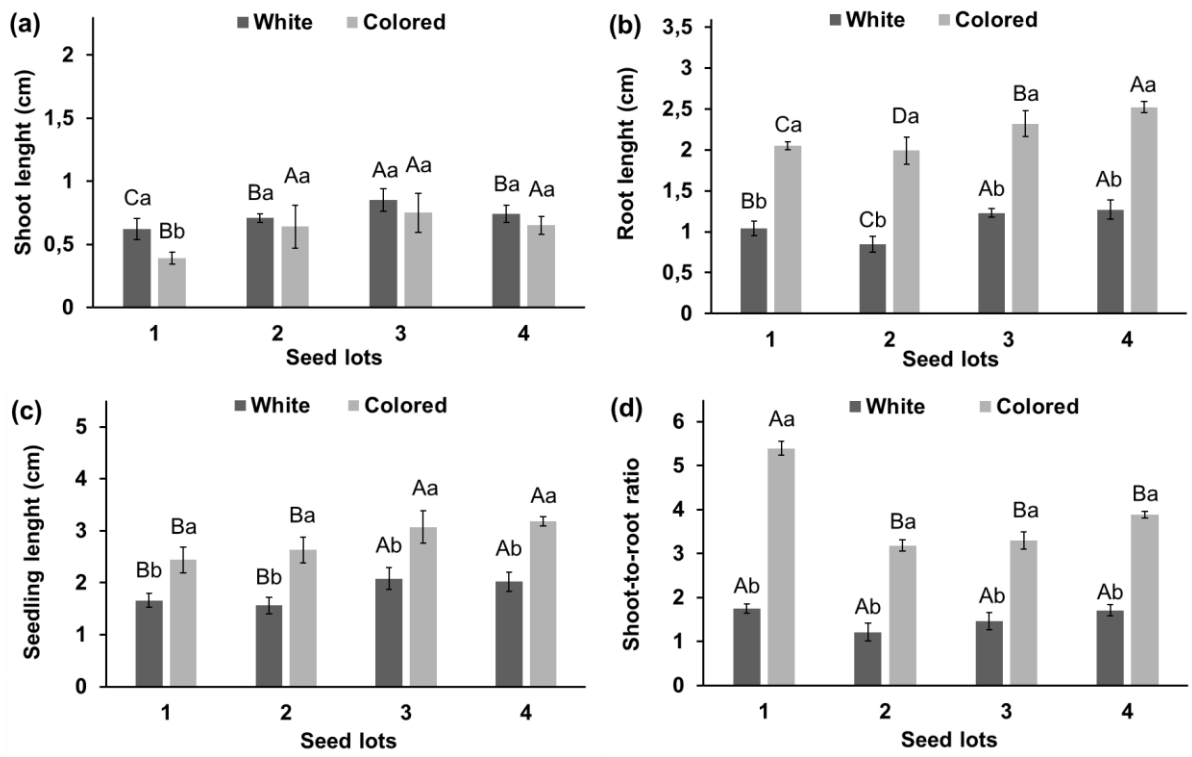


**Figure 2.** Seedling growth of *Lactuca sativa* plants under two light treatments four days after sowing, as measured by shoot (a) and root length (b), total seedling length (c), and shoot-to-root ratio (d). The means followed by equal letters, uppercase letters comparing seed lots in each light spectrum and lowercase letters comparing each lot between light spectra are not significantly different according to Tukey's test ($p \leq 0.05$).

Seedling growth was also measured seven days after sowing (Figure 3). Like those for plants after 4 days, the root and total seedling lengths as well as the shoot-to-root ratio were greater under colored light than under white light (Figures 3b, 3c, and 3d). However, unlike in plants after 4 days, in plants subjected to colored light, the shoot length was greater in lot 4 than in those subjected to white light, and no significant differences among the light treatments were observed for the remaining lots. An increase in root length decreases the shoot-to-root ratio, indicating better root support for shoot growth by providing better water and nutrient absorption (Lin *et al.*, 2013).

Root elongation under colored light at 4 and 7 days after sowing can be explained by the positive influence of blue light on root growth. Shen *et al.* (2022) reported that the root growth of Camellia sinensis was greater under blue light than under white light. Similar results were also found by Gil *et al.* (2020), who improved rooting in

Chrysanthemum plants via the combination of blue light and exogenous auxin. However, in lettuce, a combination of red and blue light also improved root growth and the antioxidant system (Bian *et al.*, 2018).

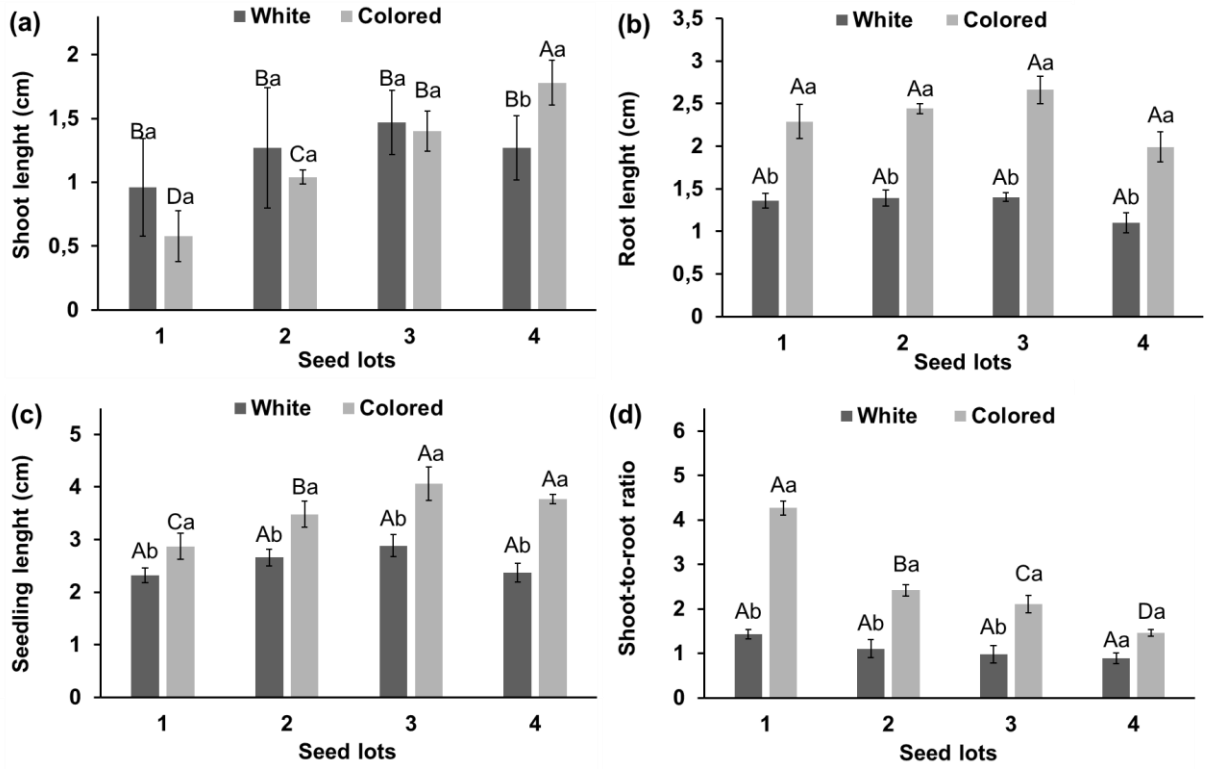


**Figure 3.** Seedling growth of Lactuca sativa plants under two light treatments seven days after sowing, as measured by shoot (a) and root length (b), total seedling length (c), and shoot-to-root ratio (d). The means followed by equal letters, uppercase letters comparing seed lots in each light spectrum and lowercase letters comparing each lot between light spectra are not significantly different according to Tukey's test ($p \leq 0.05$)

After analyzing all the results for the first phase of the experiment, it was determined that the lot that had the highest physiological quality was lot 4; therefore, this lot was chosen for the second phase of the experiment, which included physiological and biochemical analyses. Germination and GSI were measured under three light treatments (white LED, white fluorescent, and colored LED), and no significant differences were detected among the treatments (Table 2).

This may indicate that the germination of high-quality seeds is not influenced by light. However, according to the results from the first phase of the experiment and the data reported by Shimizu *et al.* (2011), lettuce plants grown under a combination of red + blue LED lights had greater rates of photosynthesis than those grown under fluorescent light compared to those grown under monochromatic lights.

In addition, under the combination of red + blue light, the length of the roots was greater than that under the other treatments. This suggested that although the germination of high-quality seeds was not influenced by light treatment, seedling growth may be greater under the combination of red + blue light.

**Table 2.** The percentage of germination and germination speed index (GSI) of lettuce seeds under three light treatments

| Light treatment | Germination (%) | GSI |
|---|---|---|
| White LED | 97,5 A** | 36,1 A |
| Fluorescent white | 97,5 A | 37,1 A |
| Colored (red + blue) LED* | 95,0 A | 39,3 A |
| **CV (%)** | 2,53% | 7,29% |

*Scheme performed according to Table 1.

**Means followed by equal letters on columns are not significantly different among light treatments according to Tukey's test (p≤ 0.05).

Although essential for photosynthesis and consequently for plant survival, light may also cause abiotic stress if these conditions are outside the tolerance range of the plant. Photosystem II (PSII) plants are vulnerable to ROS under all light conditions, especially under extreme light ratios (Murata *et al.*, 2007). Under light stress conditions, an excessive amount of accumulated energy results in ROS production in leaves (Cakmak; Kirkby, 2008), and photooxidative damage caused by ROS is responsible for chlorosis and lipid peroxidation of cellular membranes (Sevengor *et al.*, 2011). Considering this, the quantification of enzymes that are part of the antioxidant system may clarify the results until now.

The results of the quantification of SOD, CAT, and APX enzymes in seeds and seedling lettuce plants grown under the three light treatments are shown in Figure 4. SOD activity was greater in the seeds (control treatment) than in the seedlings. Light treatment did not influence the SOD activity of the lettuce plants, as there was no significant difference among the light treatments (Figure 4a). Similarly, for CAT activity, there were no significant differences among the light treatments, with seeds (control treatment) exhibiting greater CAT activity (Figure 4b). Catalase converts H2O¬2 to water and molecular oxygen (Noctor *et al.*, 2000), which is a complementary function of SOD; consequently, the results of both enzymes corroborate each other.

In contrast to those in the SOD and CAT treatments, the APC activity in the light treatment group was greater than that in the seed group (Figure 4c). This indicates that hydrogen peroxide was produced in the lettuce seedlings and was decomposed mostly by APX. This enzyme acts similarly to CAT, using ascorbate as an electron donor; however, ascorbate has a greater affinity for H2O2 than does catalase (Sharma *et al.*, 2012). Similar results were found by Zha *et al.* (2020), who reported higher APX activity in lettuce plants under high-intensity light. Light intensity directly influences ascorbate quantity, where an increase in irradiance intensity leads to an increase in ascorbate content (Dowdle *et al.*, 2007; Fukunaga*;* Fujikawa; Esaka, 2010).

The expression of the enzymes evaluated in this research corroborates the findings of previous studies showing that light is a source of stress, where the higher the light intensity and duration are, the greater the ROS production (Heyneke *et al.*, 2013). The results of the present study showed that light treatments did not cause any stress to the seedlings, indicating that analysis of the antioxidant system can be used as an indicator of stressful conditions in plants (Alscher, 2002).

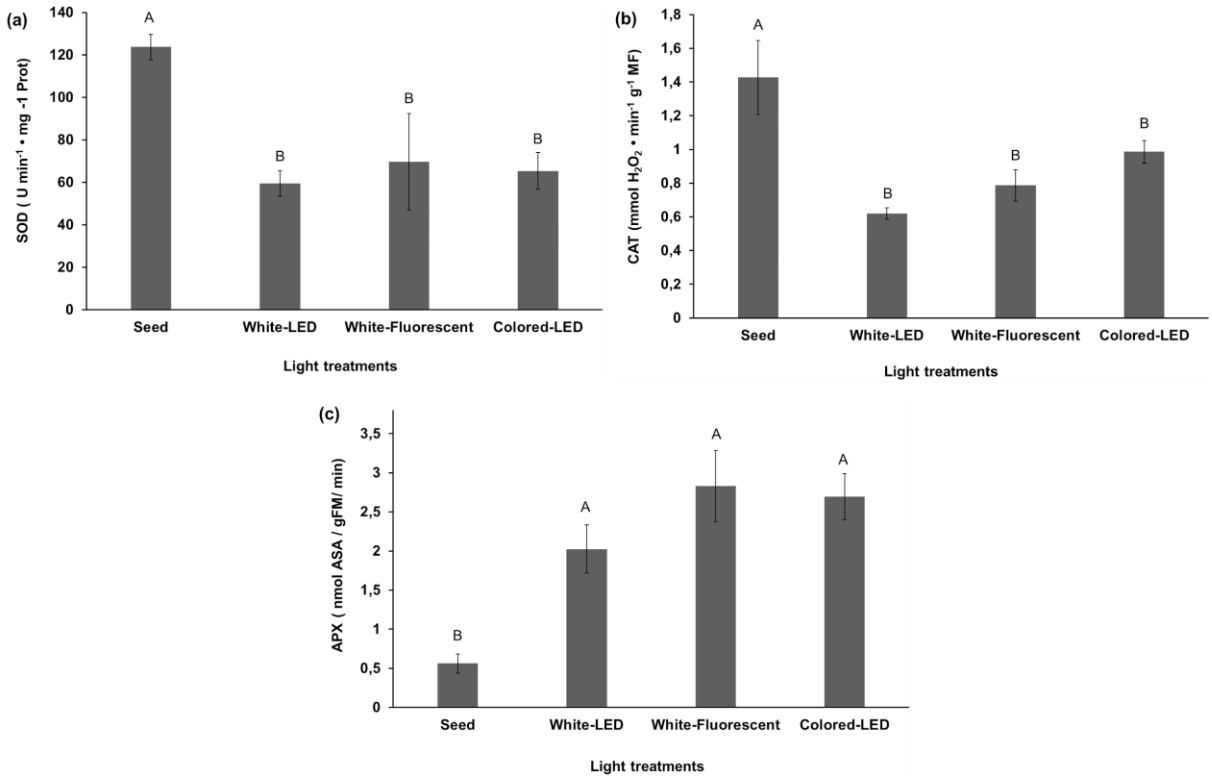


**Figure 4.** Quantification of the activity of superoxide dismutase (SOD) (a), catalase (CAT) (b), and ascorbate peroxidase (APX) (c) enzymes in seedlings developed under three light treatments and a control treatment (seeds). Means followed by equal letters are not significantly different among light treatments according to Tukey's test ($p \leq 0.05$).

Light can be a viable way to improve plant productivity (Velez-Ramirez *et al.*, 2011). The total light amount that a plant can receive within 24 hours is called the daily light integral (DLI) and is a species-related value. Constant light may excessively increase the DLI, and consequently, stress conditions will be imposed on the plant. This stressful condition results in ROS accumulation from excessive energy and reactions in chloroplasts (Heyneke *et al.*, 2013). These findings corroborate the results of our research, which indicate that no stressful conditions were applied to the plants, at least not at a level that may result in prejudices on plant growth. Additionally, in addition to the lack of differences in the light spectrum used for seed germination, seedling growth was influenced by the light, which highlights that production is affected by light and, consequently, the necessity of testing to improve profit.

## 4 CONCLUSIONS

Red light improves the speed of seed germination but not the percentage of germination.

A combination of red + blue lights stimulate lettuce seedling growth and consequently may be appropriate for seedling production.

Due to the positive effects of white light on shoot growth, the combination of white and colorful lights may be a useful option.

No photooxidative stress was observed when the plants were exposed to colored (red + blue) lights for seven days after sowing (12 h photoperiod).

## ACKNOWLEDGMENTS

The authors would like to thank the following agencies: Coordination for the Improvement of Higher Education Personnel (CAPES on the Portuguese acronym), the Minas Gerais State Research Support Foundation (FAPEMIG on the Portuguese acronym) and the National Council for Scientific and Technological Development (CNPq on the Portuguese acronym - Process 426309/2018-9). HOS is a CNPq productivity fellow (Process 310211/2021-2).

## FUNDINGS

This research was supported by the agencies Coordination for the Improvement of Higher Education Personnel (CAPES), the Minas Gerais State Research Support Foundation (FAPEMIG), and the National Council for Scientific and Technological Development (CNPq)

## REFERENCES

AN, Z. F.; ZHOU, C. J. Light induces lettuce seed germination through promoting nitric oxide production and phospholipase D-derived phosphatidic acid formation. **South African Journal of Botany**, v. 108, p. 416-422. 2017. https://doi.org/10.1016/j.sajb.2016.09.010

ALSCHER, R. G. Role of superoxide dismutases (SODs) in controlling oxidative stress in plants. **Journal of Experimental Botany**, v. 53, p. 1331–1341. 2002.https://doi.org/10.1093/jexbot/53.372.1331

BIAN, Z. *et al*. Effect of green light on nitrate reduction and edible quality of hydroponically grown lettuce (*Lactuca sativa* L.) under short-term continuous light from red and blue light-emitting diodes. **Environmental and Experimental Botany**, v. 153, p. 63–71. 2018. https://doi.org/10.1016/j.envexpbot.2018.05.010

BIEMELT, S.; KEETMAN, U.; ALBRECHT, G. Re-Aeration following hypoxia or anoxia leads to activation of the antioxidative defense system in roots of wheat seedlings. **Plant Physiology**, v. 116, p. 651–658. 1998. https://doi.org/10.1104/pp.116.2.651

BRASIL. **Regras para análise de sementes**. Ministério da Agricultura, Pecuária e Abastecimento. Secretaria de Defesa Agropecuária. Brasília: Mapa/ACS, 2009. 399 p.

CAKMAK, I.; KIRKBY, E. A. Role of magnesium in carbon partitioning and alleviating photooxidative damage. **Physiology Plant**, v. 133, p. 692–704. 2008. https://doi.org/10.1111/j.1399-3054.2007.01042.x

CHEN, X. L. *et al.* Growth and nutritional properties of lettuce affected by different alternating intervals of red and blue LED irradiation. **e**, v. 223, p. 44-52. 2017. https://doi.org/10.1016/j.scienta.2017.04.037

DOWDLE, J. *et al.* Two genes in Arabidopsis thaliana encoding GDP-L-galactose phosphorylase are required for ascorbate biosynthesis and seedling viability. **The Plant Journal.**, v. 52, p. 673-689. 2007. https://doi.org/10.1111/j.1365-313X.2007.03266.x

FERREIRA, D. F. A computer analysis system to fixed effects split plot type designs. **Revista Brasileira de Biometria**, v. 37, p. 529–535. 2019. https://doi.org/10.28951/rbb.v37i4.450

FINCH-SAVAGE, W. E.; LEUBNER-METZGER, G. Seed dormancy and the control of germination. **New Phytology.**, v. 171, p. 501–523. 2006. https://doi.org/10.1111/j.1469-8137.2006.01787.x

FRANZIN, S. M.; MENEZES, N. L.; GARCIA, D. C.; WRASSE, C. F. Methods for evaluating the physiological potential of lettuce seeds (In Portuguese). **Brasilian Seed Journal**, v. 26, p. 63–69. 2004. https://doi.org/10.1590/s0101-31222004000200009

FUKUNAGA, K.; FUJIKAWA, Y.; ESAKA, M. Light regulation of ascorbic acid biosynthesis in rice via light responsive cis-elements in genes encoding ascorbic acid biosynthetic enzymes. **Bioscience, Biotechnology, and Biochemistry,** v. 74, p. 888-891. 2010. https://doi.org/10.1271/bbb.90929

GIANNOPOLITS, C. N.; RIES, S. K. Superoxide Dismutases: II. Purification and Quantitative Relationship with Water-soluble Protein in Seedlings. **Plant Physiology**, v. 59, p. 315–318. 1977. https://doi.org/10.1104/pp.59.2.315

GIL, C. S.; JUNG, H. Y.; LEE, C.; EOM, S. H. Blue light and NAA treatment significantly improve rooting on single leaf-bud cutting of *Chrysanthemum* via upregulated rooting-related genes. **Science Horticulture**, v. 274, p. 109650. 2020. https://doi.org/10.1016/j.scienta.2020.109650

HAVIR, E. A.; MCHALE, N. A. Purification and characterization of an isozyme of catalase with enhanced-peroxidatic activity from leaves of *Nicotiana sylvestris*. **Archives of Biochemistry and Biophysics**, v. 283, p. 491–495. 1990. https://doi.org/10.1016/0003-9861(90)90672-L

HEYNEKE, E. *et al.* Dynamic compartment specific changes in glutathione and ascorbate levels in Arabidopsis plants exposed to different light intensities. **BMC Plant Biology**, v. 13. 2013. https://doi.org/10.1186/1471-2229-13-104

LIAO, P. A.; LIU, J. Y.; SUN, L. C.; CHANG, H. H. Can the adoption of protected cultivation facilities affect farm sustainability? **Sustain**, 12:1–17. 2020. https://doi.org/10.3390/su12239970

LIN, C. Blue light receptors and signal transduction. **Plant Cell**, v. 14, p. s207-S225. 2002. https://doi.org/10.1105/tpc.000646

LIN, K. H. *et al.* The effects of red, blue, and white light-emitting diodes on the growth, development, and edible quality of hydroponically grown lettuce (*Lactuca sativa* L. var. capitata). **Science Horticulture**, v. 150, p. 86–91. 2013. https://doi.org/10.1016/j.scienta.2012.10.002

LOVEGROVE, A.; HOOLEY, R. Gibberellin and abscisic acid signaling in aleurone. **Trends Plant Science**, v. 5, p. 102-110. 2000.

MAGUIRE, J. D. Speed of germination—Aid in selection and evaluation for seedling emergence and vigor. **Crop Science**, v. 2, p. 176–177. 1962. https://doi.org/10.2135/cropsci1962.0011183x000200020033x

METIVIER, J.; VIANA, A. M. The effect of long and short-day length upon the growth of whole plants and the level of soluble proteins, sugars, and stevioside in leaves of *Stevia rebaudiana* Bert. **Journal of Experimental Botany**, v. 30, p. 1211-1222. 1979

MORROW, R. C. LED lighting in horticulture. **HortScience**, v. 43, p. 1947–1950. 2008. https://doi.org/10.21273/hortsci.43.7.1947

MURATA, N.; TAKAHASHI, S.; NISHIYAMA, Y.; ALLAKHVERDIEV, S. I. Photoinhibition of photosystem II under environmental stress. **Biochimica et Biophysica Acta**, v. 1767, p. 414–421. 2007. https://doi.org/10.1016/j.bbabio.2006.11.019

NAKANO, Y.; ASADA, K. Hydrogen peroxide is scavenged by ascorbate-specific peroxidase in spinach chloroplasts. **Plant Cell Physiology**, v. 22, p. 867–880. 1981. https://doi.org/10.1093/oxfordjournals.pcp.a076232

NOCTOR, G.; VELJOVIC-JOVANOVIC, S.; FOYER, C. H.; GRACE, S. Peroxide processing in photosynthesis: Antioxidant coupling and redox signaling. **The Royal Society**, v. 355, p. 1465-1475. 2000. https://doi.org/10.1098/rstb.2000.0707

OHASHI-KANEKO, K. *et al.* Effect of light quality on growth and vegetable quality in leaf lettuce, spinach and komatsuna. **Environmental Control in Biology**, v. 45, p. 189–198. 2007. https://doi.org/10.2525/ecb.45.189

OLSZEWSKI, N.; SUN, T. P.; GUBLER, F. Gibberellin signaling: Biosynthesis, catabolism, and response pathways. **Plant Cell**, v. 14, p. 61-80. 2002. https://doi.org/10.1105/tpc.010476

PANIAGUA-PARDO, G. *et al.* Efecto de la luz led de alta intensidad sobre la germinación y el crecimiento de plántulas de brócoli (*Brassica oleracea* L.). **Polibotánica**, v. 40, p. 199-212. 2015.

PENG, J. *et al.* The Arabidopsis GAI gene defines a signaling pathway that negatively regulates gibberellin responses. **Genes & Development**, v. 11, p. 3194-3205. 1997.

SABIR, N.; SINGH, B. Protected cultivation of vegetables in global arena: A review. **Indian Journal of Agricultural Sciences**, v. 83, p. 123–135. 2013.

SABZALIAN, M. R. *et al.* High performance of vegetables, flowers, and medicinal plants in a red–blue LED incubator for indoor plant production. **Agronomy for Sustainable Development**, v. 34, p. 879–886. 2014. https://doi.org/10.1007/s13593-014-0209-6

SALA, F. C.; DA COSTA, C. P. Retrospectiva e tendência da alfacicultura Brasileira. **Horticultura Brasileira**, v. 30, p.187–194. 2012. https://doi.org/10.1590/S0102-05362012000200002

SÁNCHEZ, R. A.; SUNELL, L.; LABAVITCH, J. M.; BONNER, B. A. Changes in the endosperm cell walls of two Datura species before radicle protrusion. **Plant Physiology**, v. 93, p. 89–97.1990. https://doi.org/10.1104/pp.93.1.89

SEO, M. *et al.* Regulation of hormone metabolism in Arabidopsis seeds: phytochrome regulation of abscisic acid metabolism and abscisic acid regulation of gibberellin metabolism. **Plant Journal**, v. 48, p. 354–366. 2006. https://doi.org/10.1111/j.1365-313X.2006.02881.x

SEVENGOR, S.; YASAR, F.; KUSVURAN, S.; ELLIALTIOGLU, S. The effect of salt stress on growth, chlorophyll content, lipid peroxidation and antioxidative enzymes of pumpkin seedling. **African Journal of Agricultural Research**, v. 6, p. 4920–4924. 2011.

SINGH, D.; BASU, C.; MEINHARDT-WOLLWEBER, M.; ROTH, B. LEDs for energy efficient greenhouse lighting. **Renewable and Sustainable Energy Reviews**, v. 49, p. 139-147. 2015. https://doi.org/10.1016/j.rser.2015.04.117

SHARMA, P.; JHA, A. B.; DUBEY, R. S.; PESSARAKLI, M. Reactive oxygen species, oxidative damage, and antioxidative defense mechanism in plants under stressful conditions. **Journal of Botany**, v. 2012, p. 1–26. 2012. https://doi.org/10.1155/2012/217037

SHEN, Y. *et al.* Red and blue light affect the formation of adventitious roots of tea cuttings (*Camellia sinensis*) by regulating hormone synthesis and signal transduction pathways of mature leaves. **Frontiers Plant Science**, v. 13, p. 943662. 2022. https://doi.org/10.3389/fpls.2022.943662

SHIMIZU, H. *et al.* Light environment optimization for lettuce growth in plant factory. In: 18th **International Federation of Automatic Control (IFAC) World Congress**. IFAC Proceedings Volumes, v. 44, p. 605–609. 2011.

SOLANO, C. J. *et al.* **A LED-based smart experimental chamber to promote germination and growth of pea and melon plants: effect on the antioxidative metabolism.** In: JOAO, S. D. (ed) Prime Archives in Agricultural Research. India: Hyderabad, 2021. p. 62-83.

TAIZ, L.; ZEIGER, E.; MALLER, IM.; MURPHY, A. **Plant Physiology and Development.** 6th ed. England: Oxford University Press, 2017.

TYLER, L. *et al.* Della proteins and gibberellin-regulated seed germination and floral development in Arabidopsis. **Plant Physiology**, v. 135, p. 1008–1019. 2004. https://doi.org/10.1104/pp.104.039578

VELEZ-RAMIREZ, A. I.; VAN IEPEREN, W.; VREUGDENHIL, D.; MILLENAAR, F. F. Plants under continuous light. **Trends Plant Science**, v. 16, p. 310-318. 2011. https://doi.org/10.1016/j.tplants.2011.02.003

YANG, Y.; PEI, Q. Efficient blue–green and white light-emitting electrochemical cells based on poly[9,9-bis(3,6-dioxaheptyl)-fluorene-2,7-diyl]. **Journal of Applied Physics**, v. 81, p. 3294–3296. 1997. https://doi.org/10.1063/1.364313

YORIO, N. C. *et al.* Improving spinach, radish, and lettuce growth under red light-emitting diodes (LEDs) with blue light supplementation. **HortScience**, v. 36, p. 380–383. 2001. https://doi.org/10.21273/hortsci.36.2.380

ZHA, L. *et al.* Regulation of ascorbate accumulation and metabolism in lettuce by the red:blue ratio of continuous light using LEDs. **Frontiers in Plant Science**, v. 11, p. 704. 2020. https://doi.org/10.3389/fpls.2020.00704