

ORIGINAL RESEARCH ARTICLE

Effects of LED light intensity on purple leaf lettuce seedlings

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ABSTRACT

To explore the light intensity suitable for the growth of purple leaf lettuce seedlings in plant factories and greenhouses, the effects of light intensity of 6000, 12,000, 18,000, 24,000, and 30,000 lx on growth morphology, nutritional quality, photosynthetic pigment, and photosynthetic characteristics of purple leaf lettuce seedlings were studied with 'Beizisheng 3' as the experimental material under the condition of white light quality unchanged. The results showed that under a light intensity of 30,000 lx, purple leaf lettuce seedlings grew well, and the biomass, chlorophyll a content, chlorophyll b content, carotenoid content, total chlorophyll, soluble protein content, soluble sugar content, vitamin C content, anthocyanin content, net photosynthetic rate, transpiration rate, and stomatal conductance were the largest, while the nitrate content and intercellular CO₂ concentration were the lowest. However, chlorophyll b content, carotenoid content, and transpiration rate had no significant difference compared with those under 24000 lx light intensity, and the plant height was the highest under 24,000 lx light intensity. Under the condition of white light quality, the most suitable light intensity for the growth of purple leaf lettuce seedlings is 30,000 lx.

Keywords: light intensity; purple leaf lettuce seedlings; growth morphology; nutritional quality; photosynthetic pigment; photosynthetic characteristics

1. Introduction

Light is a key environmental factor involved in plant growth and development. It not only supplies energy for plant photosynthesis but also regulates many signal reactions^[1] and enzyme activities^[2] in plants. The light environment mainly includes three aspects: light intensity (light quantity), light quality (spectral distribution) and light period (light duration)^[3]. In addition to light quality and light period, light intensity also has a significant impact on plant nutritional growth, reproductive growth, photosynthesis, physiological and biochemical activities^[4]. At present, with the restriction of facility environment on light and the frequent occurrence of adverse environmental climate, the phenomenon of weak light is becoming more and more common, which has a certain adverse impact on agricultural production and urban greening^[5]. Therefore, people's awareness of changing the light environment has been gradually strengthened and applied to the agricultural field.

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Lettuce is an herb of compositae family, which has rich nutritional value, is rich in protein, vitamin C, and other nutrients, and has a variety of health functions such as anti-aging, prevention of arrhythmia, prevention of cancer cell formation. It has won the favor of most consumers^[6]. According to leaf color, it can be divided into green leaf lettuce and purple leaf lettuce^[7]. Compared with ordinary green leaf lettuce, purple leaf lettuce has a higher content of total phenols, anthocyanins, vitamins, etc. It not only has strong antioxidant capacity, but also can improve vision, prevent eye fatigue, etc. It is popular with the public with its unique leaf color form, and has good market value^[8].

As people have higher and higher requirements for vegetable quality, traditional open field cultivation and solar greenhouse cultivation can no longer meet the demand^[9]. At present, LED lights have been used in most artificial light plant factories, and LED intelligent plant factories are the development direction of plant factories^[10]. LED is a new type of semiconductor energy-saving light source. Compared with traditional artificial light sources such as incandescent lamps, LED has the characteristics of monochromatic light, small size, good controllability, long service life, etc., which largely makes up for the shortcomings of traditional light sources in facility agricultural production, and has more obvious advantages in plant supplementary lighting^[11]. Therefore, the application of LED light sources as a light supplement facility has continuously developed into a new agricultural model.

Research results at home and abroad have shown that lettuce μ It has the largest dry biomass under high illumination of 220 μ mol/(m²·s) and low nitrogen of 7 mmol/L, and is conducive to promoting the increase of vitamin C content and the decrease of nitrate content. Compared with high illumination, the nitrogen supply under low illumination has a more obvious impact on chlorophyll concentration^[12]. Viršilė et al.^[13] compared the effects of different light intensities and photoperiods on the growth and nitrate assimilation of red and green leaf lettuce (lettuce). The results show that 300~400 μ Mol/(m²·s)light intensity and 16–18 h photoperiod are necessary for red and green leaf lettuce, and light intensity is more conducive to nitrate assimilation than photoperiod. Ren et al.^[14] showed that under the condition that the light quality was red light: blue light = 1:1, the nutritional quality of purple lettuce increased with the increase of light intensity, and the increase of appropriate light intensity could promote the increase of biomass in purple lettuce. With the progress of science and technology, the research on light intensity for lettuce is constantly developing, which is of great significance for cultivating strong seedlings in facilities and reducing light loss.

Most studies focus on the effects of different light quality or light intensity on plants, as well as the effects of different light quality on lettuce^[15], tomato^[16] and other seedlings, while there are few reports on the effects of light intensity on vegetable seedlings in plant factories and greenhouses. In this experiment, the purple loose leaf lettuce 'Beizisheng 3' was used as the experimental material to study the effects of different light intensity on the growth morphology, nutritional quality, photosynthetic pigment and photosynthetic characteristics of purple leaf lettuce seedlings by regulating the light conditions at the seedling stage, in order to screen LED light mode suitable for lettuce seedlings in artificial light plant factories and greenhouses, It provides scientific basis for the later study of the effects of different light patterns on the growth of plant seedlings.

2. Materials and methods

2.1. Test materials

The test material was 'Beizisheng No. 3', which was selected by Beijing Agricultural University and was a purple loose leaf lettuce.

2.2. Test method

The experiment was conducted on 6 December 2021, in the High Quality and Safe Vegetable Cultivation Technology Laboratory of Beijing Agricultural University. The purple leaf lettuce seeds were planted in a plastic two-color basin after germination and whitening, and were treated with five different intensities of white LED lights. See **Table 1** for each treatment. The light incubator (model PRX–450D) of Ningbo Saifu Co., Ltd. is used. The temperature of the light incubator in the day and night is (22 ± 2) °C/(15 ± 2) °C, the relative humidity is (60 ± 10) %, the light cycle is set to 14 h/d, and water is poured every 3 days. 60 seedlings from each treatment were collected after 25 days. During the collection, 3 seedlings of purple leaf lettuce were randomly selected from each treatment for data measurement, and each treatment was repeated three times.

Table 1. LED light intensity of different treatments.			
Treatment	Light intensity/lx		
T1	6000		
T2	12000		
T3	18000		
T4	24000		
T5	30000		

Table 1. LED light intensity of different treatments

2.3. Project measurement

The leaf length, leaf width, petiole length, plant height, spread and root length of lettuce seedlings were measured with ruler and tape. The stem diameter is measured with vernier caliper. The above ground fresh weight, the underground fresh weight, the above ground dry weight, and the underground dry weight were measured with a 0.0001 g analytical balance, and each treated sample was put into an oven at 105 °C for 15 min, and then dried at 80 °C to a constant weight for weighing. The number of blades is counted by direct method. The calculation results for leaf area, root shoot ratio and healthy seedling index are as shown in Equations (1)~(3).

$$Leaf area = leaf length \times Leaf width \times 0.7007$$
(1)

Root shoot ratio =
$$\frac{\text{fresh weight of underground}}{\text{Fresh weight above ground}}$$
 (2)

Healthy seedling index = dry weight of the whole plant ×
$$\left(\frac{\text{Stem diameter}}{\text{Plant height}} + \frac{\text{underground dry mass}}{\text{Dry mass above ground}}\right)$$
 (3)

The soluble protein content of lettuce seedlings was determined by Coomassie Brilliant Blue G-250 staining method. The content of soluble sugar was determined by spectrophotometry. The content of vitamin C was determined by ELISA. The anthocyanin content of leaves was determined by visible spectrophotometry. The nitrate content was determined by ultraviolet spectrophotometry. Chlorophyll and carotenoid content were determined by 95% ethanol extraction method. CIRAS–3 portable photosynthetic instrument (Lufthansa Scientific Instrument Co., Ltd.) was used to measure the net photosynthetic rate, intercellular CO₂ concentration, transpiration rate and stomatal conductance.

2.4. Data analysis

Microsoft Excel 2007 and SPSS 17.0 software were used for statistical analysis of data, and multiple comparative analysis (Duncan) was used for significant difference analysis (P < 0.05).

3. Results and analysis

3.1. Effect of light intensity on growth morphology of lettuce seedlings

3.1.1. Seedling growth

In order to explore the growth of lettuce seedlings under different light intensities, the plant height, spread, stem diameter, petiole length, leaf length, leaf width, leaf area, leaf number and root length were measured and compared. It can be seen from **Table 2** that the growth of purple leaf lettuce seedlings under different treatments has significant differences. With the increase in light intensity, the overall spread, stem diameter, leaf length, leaf width, leaf area, leaf number and root length have increased. The plant height of T4 treatment was significantly higher than that of other treatments. The spread, stem diameter, leaf length, leaf area, leaf number, and root length of T5 treatment were significantly different from those of other treatments, but the number of leaves was not significantly different from that of T4 treatment. There was no significant difference in plant height, spread, leaf length, leaf area, leaf number and root length of T2 treatment was significantly higher than that of T1, T3, T5 treatment, but there was no significant difference with T4 treatment. It can be seen that increasing the light intensity within a certain range is beneficial to the growth of lettuce seedlings.

Table 2. Effect of light intensity on the growth of purple fettuce seedings.					
Growth index	T1	T2	Т3	T4	Т5
Plant height/cm	$4.07 \pm 0.06 cd$	$4.43\pm0.31c$	$3.83 \pm 0.06 d$	$7.70\pm0.35a$	$6.27\pm0.06b$
Spread/cm	$8.17\pm0.06d$	$8.47\pm 0.12d$	$9.27\pm0.15\text{c}$	$14.47\pm0.06b$	$15.70\pm0.56a$
Stem diameter/mm	$1.83\pm0.29\text{c}$	$1.88\pm0.11\text{c}$	$2.17\pm0.29c$	$3.33\pm0.58b$	$4.33\pm0.58a$
Petiole length/cm	$1.13\pm0.29\text{b}$	$1.47\pm0.06a$	$0.97\pm0.15b$	$1.23\pm0.12ab \\$	$0.67\pm0.06\texttt{c}$
Leaf length/cm	$4.20\pm0.10d$	$4.47\pm0.15d$	$5.00\pm0.10\text{c}$	$7.40\pm0.20b$	$7.87 \pm 0.21 a$
Leaf width/cm	$2.23\pm0.12e$	$2.53\pm0.15\text{d}$	$3.40\pm0.20\text{c}$	$6.13 \pm 0.06b$	$6.40\pm0.10a$
Leaf area/cm ²	$6.57\pm0.38d$	$7.93\pm 0.52d$	$11.92\pm0.84c$	$31.80\pm0.62b$	$35.28 \pm 1.19a$
Number of leaves/sheet	$4.00\pm0.00\text{c}$	$4.00\pm0.00\text{c}$	$5.00\pm0.00b$	$8.00\pm0.00a$	$8.33\pm0.58a$
Root length/cm	$9.23 \pm 1.36 d \\$	$9.60\pm0.52d$	$11.13\pm0.76c$	15.20±0.70b	$18.33\pm0.58a$

Table 2. Effect of light intensity on the growth of purple lettuce seedlings

Note: Different letters in the same line indicate significant difference between treatments (P < 0.05), the same below.

3.1.2. Seedling biomass

In order to explore the differences of biomass of lettuce seedlings under different light intensities, the above ground fresh weight, above ground dry weight, underground fresh weight, underground dry weight, root shoot ratio (fresh weight) and strong seedling index were measured and compared. It can be seen from **Table 3** that with the increase in light intensity, the aboveground fresh weight, aboveground dry weight, underground fresh weight, underground dry weight, root shoot ratio and strong seedling index of purple leaf lettuce seedlings show an upward trend. The fresh weight and root shoot ratio under T5 treatment were significantly different from the other treatments, but there was no significant difference between the other treatments. Compared with other treatments, the fresh weight and strong seedling index of shoot under T5 treatment were significantly different, but there was no significant difference among T1, T2 and T3 treatments. Therefore, T5 treatment is beneficial to increase the biomass of lettuce seedlings.

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Biomass index	T1	T2	Т3	T4	Т5
Fresh weight of shoot/g	$0.30\pm0.05\text{c}$	$0.44\pm0.11\text{c}$	$0.60\pm0.06c$	$4.48\pm0.61\text{b}$	$5.76\pm0.86\ a$
Underground fresh weight/g	$0.02\pm0.01\text{b}$	$0.03\pm0.02b$	$0.05\pm0.01b$	$0.36\pm0.15\text{b}$	$1.39\pm0.40a$
Dry weight above ground/mg	$15.77\pm0.45d$	$16.47\pm0.06d$	$30.13\pm0.12c$	$257.40\pm0.20b$	$336.53\pm0.91a$
Underground dry weight/mg	$2.40\pm0.10e$	$3.37\pm 0.23d$	$5.83\pm0.29\text{c}$	$25.60\pm0.50b$	$81.87\pm0.32a$
Root shoot ratio (fresh weight)	$0.06\pm0.02b$	$0.08\pm0.02b$	$0.08\pm0.01b$	$0.08\pm0.02b$	$0.24\pm0.06a$
Robust seedling index	$0.01\pm0.00\text{c}$	$0.01\pm0.00\text{c}$	$0.03\pm0.00\text{c}$	$0.15\pm0.02b$	$0.39\pm0.04a$

Table 3. Effect of light intensity on biomass of lettuce seedlings.

3.2. Effect of light intensity on the nutritional quality of lettuce seedlings

In order to explore the difference in nutritional quality of purple leaf lettuce seedlings under different light intensities, five indexes were measured and compared. It can be seen from **Figure 1** that the nutritional quality of purple leaf lettuce seedlings is improved with the increase in light intensity, and the contents of soluble protein, vitamin C, soluble sugar and anthocyanin are increasing. Under T5 treatment, the content of soluble protein, vitamin C, soluble sugar and anthocyanin in purple leaf lettuce seedlings showed the highest value, but the content of soluble protein had no significant difference compared with T4 treatment. Under T1 treatment, the other quality indexes except nitrate content were the minimum values.



Figure 1. Effect of light intensity on nutritional quality of purple leaf lettuce seedlings. Different letters indicate significant difference between treatments (P < 0.05), the same below.

To sum up, the contents of soluble protein, soluble sugar, vitamin C and anthocyanin under T5 treatment reached the maximum, the nitrate content was the minimum, and the nutritional quality was T5 > T4 > T3 > T2 > T1. Therefore, the increase in light intensity can gradually improve the nutritional quality of lettuce seedlings.

3.3. Effect of light intensity on photosynthetic pigment of purple leaf lettuce seedlings

In order to explore the difference in photosynthetic pigment content of purple leaf lettuce seedlings under different light intensities, the photosynthetic pigments were measured and compared. It can be seen from **Table 4** that under T5 treatment, the chlorophyll a, chlorophyll b, carotenoid content and total chlorophyll content of purple leaf lettuce seedlings reach the maximum, which is significantly different from other treatments, but the chlorophyll b content and carotenoid content have no significant difference compared with T4 treatment, and the photosynthetic pigment content of seedlings under T1 treatment is the lowest. It indicated that T5 treatment significantly increased the contents of chlorophyll a, chlorophyll b, carotenoids and the total amount of chlorophyll in lettuce seedlings, and accelerated the synthesis of photosynthetic pigments to facilitate energy conversion.

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Pigment	T1	T2	Т3	T4	Т5
Chlorophyll a	$0.98\pm0.05d$	$1.18\pm0.08\text{c}$	$1.21\pm0.05\texttt{c}$	$1.66\pm0.04b$	$1.88\pm0.06a$
Chlorophyll b	$0.40\pm0.00\text{c}$	$0.43\pm0.05\text{c}$	$0.48\pm0.02bc$	$0.54\pm0.08ab$	$0.57\pm0.03a$
Carotenoids	$0.57\pm0.04c$	$0.74\pm0.07b \\$	$0.78\pm0.06b$	$1.10\pm0.06a$	$1.21\pm0.10a$
Total chlorophyll	$1.38\pm0.05d$	$1.61\pm0.12\text{c}$	$1.69\pm0.06\text{c}$	$2.20\pm0.12b$	$2.45\pm0.06a$

Table 4. Effect of light intensity on photosynthetic pigment content of purple leaf lettuce seedlings mg/g.

3.4. Effect of light intensity on photosynthetic characteristics of lettuce seedlings

In order to explore the differences in photosynthetic characteristics of lettuce seedlings under different light intensities, four basic photosynthetic parameters were measured and compared. It can be seen from **Figure 2** that the transpiration rate, net photosynthetic rate and stomatal conductance of purple leaf lettuce seedlings gradually increase with the increase in light intensity, and the intercellular CO_2 concentration decreases with the increase in light intensity. The net photosynthetic rate, stomatal conductance and transpiration rate reached their highest values under T5 treatment, but the transpiration rate had no significant difference compared with T4 treatment; Under T1 treatment, the intercellular CO_2 concentration of lettuce seedlings was the highest, while under T5 treatment, the intercellular CO_2 concentration was the lowest. Therefore, T5 treatment was beneficial to the improvement of photosynthetic characteristics of lettuce seedlings.



Figure 2. Effect of light intensity on photosynthetic characteristics of purple leaf lettuce seedlings.

4. Conclusion

30,000 lx treatment can promote the growth and biomass increase of purple leaf lettuce seedlings. Under this light intensity, the soluble protein content, soluble sugar content, vitamin content and anthocyanin content of purple leaf lettuce seedlings are the highest and the nitrate content is the lowest, which can promote the accumulation of photosynthetic pigments and improve the photosynthetic characteristics. This light intensity can be selected as the light intensity of seedling breeding according to the breeding environment.

5. Discussion

Various forms of plants are the process of plants adapting to light intensity. Strong light and weak light will inhibit the growth process of plants to some extent^[17]. Weak light will lead to excessive growth and thin leaves of plants, while strong light will lead to water loss and wilting of plants, small and yellow leaves, etc.^[18]. Liu et al.^[17] studied the effect of light intensity on cherry tomato seedlings. μ Under mol/(m²·s) light intensity, the plant has a good growth trend, which is suitable for cultivating cherry tomato seedlings. Based on the experience of predecessors, this experiment conducted a light intensity test on purple leaf lettuce seedlings. The results showed that, with the increase of light intensity, the plant height, spread, stem diameter, leaf length, leaf width, leaf area, number of leaves and root length of purple leaf lettuce seedlings increased in general, while the petiole length decreased, which was considered to be due to the phenomenon of excessive length of seedlings under weak light. With the increase of light intensity, the fresh weight above ground, fresh weight below ground, dry weight above ground, dry weight below ground, root shoot ratio and healthy seedling index of purple leaf lettuce seedlings increased, which is consistent with the test results of Jiang et al.^[19]. It may be caused by the small range of light intensity or the seedling stage. Therefore, the increase in light intensity in a certain range is beneficial to the growth and biomass of lettuce seedlings.

Light intensity affects the nutritional quality of plants. Too weak light intensity is likely to lead to the decline of plant nutritional quality. The quality of vegetable can be judged by the content of soluble protein, anthocyanin, nitrate, etc. Among them, soluble protein is an important nutrient in vegetables, providing protein and energy for the human body; soluble sugar can also provide energy for human life activities; vitamin C has

an anti-aging protective function; anthocyanins have many effects, such as resisting oxidation and inflammation; The advantages and disadvantages of nitrate content are still controversial. An appropriate amount of nitrate is harmless to human body, while excessive nitrate is harmful to human digestive system^[20]. The research results of Ji et al.^[21] show that the increase in light intensity is conducive to the increase of soluble protein and the accumulation of sugar, thus improving the quality of lettuce. The results of this experiment show that with the increase in light intensity, the contents of soluble protein, soluble sugar, vitamin C and anthocyanin in purple leaf lettuce seedlings show an increasing trend, while the nitrate content shows a decreasing trend, and the content of other indicators reaches the highest at 30000 lx, except nitrate content, because higher light intensity improves its nitrate reductase activity and reduces the accumulation of nitrate^[22]. The results of this experiment are consistent with those of Wang et al.^[23]. Increasing the appropriate light intensity will help to improve the nutritional quality of lettuce.

The organic matter produced by plant photosynthesis mainly supplies it for life activities, and the light intensity also affects the intensity of plant photosynthesis and the rise and fall of photosynthetic pigments^[24]. Kwack et al.^[25] showed that increasing light intensity can promote photosynthesis in plants and increase their carbohydrate accumulation. The results showed that the light intensity of 30,000 lx was beneficial to the increase of chlorophyll content and carotenoid content of purple leaf lettuce seedlings; with the increase of light intensity, the net photosynthetic rate, stomatal conductance and transpiration rate of lettuce seedlings also showed an upward trend, while the intercellular CO₂ concentration showed an opposite trend. This test result is consistent with that of Ren et al.^[14]. Higher light intensity is conducive to an increase in photosynthetic pigments and photosynthetic characteristics. It can be seen that the moderate increase in light intensity is conducive to promoting the accumulation of photosynthetic pigments and energy conversion.

To sum up, light intensity has a significant impact on the growth and quality of lettuce seedlings. A proper increase in light intensity within a certain range is conducive to promoting the growth, quality improvement, photosynthetic pigments and photosynthetic characteristics of lettuce seedlings. The growth of plant seedlings will be affected by many environmental factors, including light, temperature, soil, etc. In this study, although a uniform light intensity gradient has been set to study its impact on lettuce seedlings, the research factors are relatively simple, and the next step needs to carry out relevant research on light quality, light cycle, temperature and other environmental factors, so as to obtain a better breeding environment, It provides a reference for future seedling raising in plant factories and greenhouses.

Conflict of interest

The authors declare no conflict of interest.

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