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Research Article

Blue Light-Emitting Diode Lighting Improves Antioxidant Potential in Barley Germinated Seeds

Manabu Sugimoto*

Institute of Plant Science and Resources, Okayama University, Kurashiki, Okayama, Japan

ABSTRACT

Barley, a key ingredient in beer, has recently garnered attention for its potential health benefits. Specifically, barley grain has been found to promote the growth of beneficial intestinal microorganisms, positioning it as a functional food. To enhance the functional biomolecules in barley seeds, barley was germinating under Light-Emitting Diode (LED) lighting. After 4 days of germination, the barley exposed to red LED lighting displayed longer shoot elongation, higher α-amylase activity, and lower starch content compared to those germinated in the dark. On the other hand, blue LED lighting resulted in similar root and shoot elongation as the dark condition but significantly increased vitamin C content and DPPH radical scavenging activity in the germinated barley. Interestingly, red LED lighting did not show any significant effects on these antioxidant components. These findings suggest that blue LED lighting could be a potential method to produce functional germinated barley with enhanced antioxidant potential while maintaining the quality standards of malt.

Keywords: Antioxidant; Barley; Germination; LED

INTRODUCTION

Light plays an important role in regulating various morphological and physiological changes in mature plants [1,2]. Blue and red wavelengths are the primary spectral wavelengths that significantly influence the primary and secondary metabolism of plants [3]. Light-Emitting Diodes (LEDs) have been increasingly used as artificial light sources for plant growth in controlled environments. Research has shown that antioxidant activity increases in lettuce and *Gynura* when exposed to blue LED light [4,5]. Additionally, rose, *Chrysanthemum*, and *Campanula* exhibited higher flavonoid and phenolic contents when exposed to mix red and blue LED light with a higher proportion of blue [6]. Tomato seedlings cultivated under blue LED light displayed the highest antioxidant activity [7], whereas pea seedlings grown under red LED light showed the highest antioxidant potential [8]. These findings suggest that LED lighting can be strategically used to produce 'functional food'-food that contains enhanced levels of health-promoting or disease risk-reducing compounds such as antioxidants, minerals, and vitamins. However, it is important to note that the effects of LED lighting vary depending on the plant cultivar.

Barley is a versatile crop with multiple applications, serving as a source of food, livestock feed, and an essential ingredient for malt brewing. Recently, there has been growing interest in barley as a potential functional food. Young green barley leaves are particularly rich in antioxidants, flavones C-glycoside, saponarin, lutonarin, minerals, and vitamins, which are associated with various physiological benefits [9-12]. Additionally, barley grain contains β-glucan, a type of soluble dietary fiber [13,14] that has been linked to positive effects on obesity, cardiovascular diseases, diabetes, and cholesterol levels [15-17]. It also promotes the growth of beneficial intestinal microorganisms, such as lactobacilli and bifidobacteria [18-20]. Previous studies on the influence of LED light on young green barley leaves found that red LED light promoted plant height and weight gain [21], while blue LED light increased the contents of saponarin and polyphenol compared to red or far-red LED lighting [22]. However, the effects of LED lighting on the quality of germinated barley seeds remain unclear.

Correspondence to: Manabu Sugimoto, Institute of Plant Science and Resources, Okayama University, Kurashiki, Okayama, Japan, Telephone: +81 86 434 1253; E-mail: manabus@okayama-u.ac.jp

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In the present study, barley seeds were germinated under LED lighting to enhance their quality. The results showed that the seeds germinated under blue LED light for 4 days exhibited increased levels of vitamin C and DPPH radical scavenging activity. On the other hand, seeds germinated under red LED light displayed higher α-amylase activity, lower starch content, and longer shoot length compared to seeds germinated in dark.

MATERIALS AND METHODS

Germination and light treatment conditions

Seeds of malting barley, *Hordeum vulgare* L., Haruna Nijo, were soaked in water at 15°C for 5 h. The seeds were then arranged in a single layer on a water-filled sponge. They were incubated at 20°C for 4 days under constant lighting conditions: Red LED light (660 nm, 140 µmol/m^2/sec), blue LED light (470 nm, 140 µmol/m^2/sec), or kept in darkness. After germination, the shoots and roots were removed, and the seeds were stored at -80°C for further analysis.

Preparation of extracts

Five germinated seeds were ground into a fine powder using a mortar and pestle, and this powder was considered one sample. The weight of the powder was recorded, and it was then transferred to a glass centrifuge tube. The powder was suspended in 3 ml of 80% methanol for total polyphenol content and DPPH radical scavenging activity assays. For the vitamin C assay, the powder was suspended in 3 ml of 5% metaphosphoric acid. The suspensions were incubated with shaking at room temperature for 10 minutes, and then the supernatants were collected by centrifugation to be used as extracts for subsequent assays.

-Amylase activity

α-Amylase activity was measured were performed using amylase assay kit (Abcam, Cambridge, UK) according to the manufacturer's protocol. The rate of p-nitrophenol formation was determined by measuring at 405 nm.

Starch assay

The starch content in the germinated seeds was assayed using Starch Assay Kit (Cell Biolabs, INC., San Diego, USA) according to the manufacturer's protocol. Twenty mg of germinated seeds was grinded to prepare the starch extract. The extract diluted 100 times was used to the assay.

Total polyphenol assay

The total polyphenol content in the extracts was analyzed using the Folin-Ciocalteu reagent method [23]. The extract was diluted ten times with water, and 0.2 ml of this diluted extract was mixed with 5 ml of Folin and Ciocalteu reagent (1:1 diluted with water). After a 3 minute incubation, 5 ml of 0.4 M Na_2CO_3 was added to the mixture. The solution was incubated at 50°C for 5 minutes and then cooled on ice. The absorbance was measured at 765 nm, and the results were compared to a standard curve prepared using tannic acid.

DPPH radical scavenging activity

The antioxidant activity of the extract was assessed using the DPPH method [24]. One ml of the extract was combined with 9 ml of 0.5 mM DPPH dissolved in 50% methanol and incubated in the dark at room temperature for 10 minutes. The decrease in absorbance was measured at 517 nm, and the results were compared to a standard curve prepared using Trolox.

Vitamin C assay

The vitamin C content was determined following the method described by Bradley et al., [25]. The extract (200 µl) was mixed with 12 µl of 3% bromine and incubated at room temperature for 5 minutes. Then, 70 µl of 5% metaphosphoric acid/2% SnCl2 solution was added, followed by 20 µl of 2% DNPH dissolved in 44% H_2SO_4). The mixture was incubated at 37°C for 3 h. Finally, 100 µl of 85% H2SO4 was added, and the solution was allowed to stand at room temperature. After 30 min, the absorbance was measured at 530 nm, and the results were compared to a standard curve prepared using L-ascorbic acid.

RESULTS AND DISCUSSION

Effect of red and blue LEDs on root and shoot elongation

Red LED lighting significantly promoted the elongation of both roots and shoots in barley seeds, while blue LED lighting had no significant effect compared to the dark condition (Figure 1).

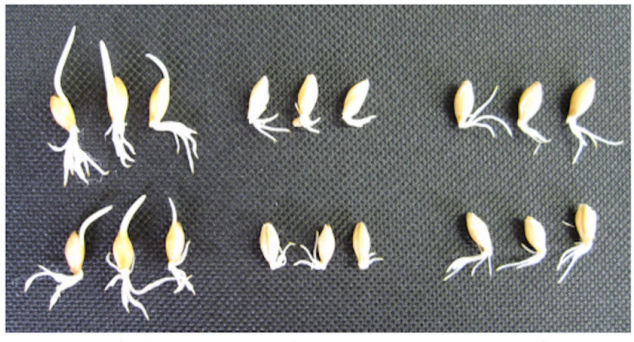

Figure 1: Barley seeds germinated for 4 days under constant lighting of red LED, blue LED, or dark conditions. Seeds were incubated at 20°C for 4 days under constant lighting conditions: red LED light (660 nm, 140 µmol/m^2/sec), blue LED light (470 nm, 140 µmol/m^2/sec), or in the dark after soaking in water at 15°C for 5 h.

The shoot length of seeds germinated for 4 days under red LED, blue LED, and dark conditions were 22.1 ± 2.9*, 10.4 ± 2.7, and 10.0 ± 2.2 mm, respectively (n=30, *p<0.001). Similarly, the root

lengths were 21.1 ± 3.9, 16.1 ± 2.3, and 19.0 ± 4.3 mm, respectively (n=30). Red LED lighting increased shoot length significantly by approximately 2.2-fold compared to the dark condition, while blue LED lighting had no significant impact. These results align with the findings reported by Kochetova et al., [26] where red LED lighting increased shoot length by 13% in 9-day-old seedlings.

In addition to the morphological changes, we also observed differences in α-amylase activities and starch content (Table 1).

Seeds	Red LED	Blue LED	Dark
α-amylase (mU/g seed	11.1 ± 0.8**	8.75 ± 0.9	8.55 ± 1.1
Starch (g/g seed)	412 ± 45*	441 ± 41	469 ± 41
DPPH radical scavenging activity (mM/g seed)	3.30 ± 0.15	3.72 ± 0.04*	3.30 ± 0.16
Total polyphenol (g/g seed)	14.1 ± 0.30	14.6 ± 1.0	14.1 ± 0.32
Vitamin C (g/g seed)	46.5 ± 1.6	78.1 ± 1.2*	55.0 ± 2.9

Note: n=3; *p<0.001 and **p<0.05

Table 1: Properties of barley seeds germinated for 4 days under constant lighting of red LED, blue LED or dark conditions.

Red LED lighting increased α-amylase activity by 1.3-fold and decreased starch content significantly compared to the dark condition. As explained by Beck et al., Fincher [27,28], α-amylase plays an important role during cereal germination, converting starch in the endosperm into nutrients that support seedling growth. The increased α-amylase activity and decreased starch content under red LED lighting suggests enhanced metabolism, which may contribute to the accelerated shoot growth observed in our study.

Effect of red and blue LEDs on antioxidant properties

Previous studies have reported conflicting results regarding the effect of LED lighting on the antioxidant properties of barley. Lee et al., found that young barley leaves grown under blue LED lighting showed an increasing tendency in antioxidant activity compared to those grown in the dark [29]. On the other hand, Paulickova et al., observed that barley sprouted for 20 days had the highest vitamin C level, while Koga et al., reported decreased vitamin C and E contents in 15-day-old barley leaves under red LED lighting compared to natural light [30,21]. These discrepancies suggest that the impact of LED lighting on antioxidant production depends on the harvest period and environmental factors.

In this study, the effect of LED lighting on DPPH scavenging activity, total polyphenol content, and vitamin C content in 4-day-old germinated barley were investigated (Table 1). The results indicate that blue LED lighting increased the levels of DPPH radical scavenging activity and vitamin C significantly. Notably, vitamin C content increased by 1.4-fold under blue LED lighting compared to the dark condition. However, red LED lighting had no significant effect on these antioxidant properties in 4-day-old germinated barley.

The malting process for brewing beer involves steeping, germinating, and kilning. Germination initiates the degradation and modification of cell wall, protein, and starch compounds, producing bioactive molecules that influence beer quality. Therefore, maintaining the quantity and quality of these molecules during germination is crucial in the malting and brewing industries. Additionally, antioxidants in germinated barley contribute to the oxidative stability and flavor of beer and offer potential health benefits for consumers by neutralizing Reactive Oxygen Species (ROS) associated with various diseases [31]. The finding that blue LED lighting, when applied for 4 days, does not affect root and shoot elongation supposes the same biomolecule content such as starch and protein, which ensures the quality of malt, and that increases the antioxidant content of germinated barley suppose the enhancing function of the germinated barley. This presents an opportunity to develop a low-cost process for producing functional germinated barley, not only as a raw material for brewing but also as an ingredient in functional foods, providing high levels of bioactive compounds with potential health-promoting properties.

CONCLUSION

The result of this study demonstrated that germination of barley seeds under constant blue LED lighting for 4 days enhances the bioactive compounds with the same quality as the germinated barley under dark condition. Blue LED lighting did not affect starch content, and α-amylase activity but enhanced DPPH radical scavenging activity and vitamin C content in the germinated barley. Thus, barley germinated barley under constant blue LED lighting for 4 days can be a valuable raw material and functional food.

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REFERENCES

1. Drozdova IS, Bondar VV, Bukhov NG, Kotov AA, Kotova LM, Maevskaya SN, et al. Effects of light spectral quality on morphogenesis and source-sink relations in radish plants. Russ J Plant Physiol. 2001;48:415-420.
2. Li Q, Kubota C. Effects of supplemental light quality on growth and phytochemicals of baby leaf lettuce. J Sci Food Agric. 2009;67(1):59-64.
3. Johkan M, Shoji K, Goto F, Hashida SN, Yoshihara T. Blue light-emitting diode light irradiation of seedlings improves seedling quality and growth after transplanting in red leaf lettuce. HortScience. 2010;45(12):1809-1814.

4. Son KH, Oh MM. Leaf shape, growth, and antioxidant phenolic compounds of two lettuce cultivars grown under various combinations of blue and red light-emitting diodes. HortScience. 2013;48(8):988-995.
5. Ren J, Guo S, Xu C, Yang C, Ai W, Tang Y, et al. Effects of different carbon dioxide and LED lighting levels on the anti-oxidative capabilities of Gynura bicolor DC. Adv Space Res. 2014;53(2):353-361.
6. Ouzounis T, Fretté X, Rosenqvist E, Ottosen CO. Spectral effects of supplementary lighting on the secondary metabolites in roses, *Chrysanthemums*, and *campanulas*. J Plant Physiol. 2014;171(16): 1491-1499.
7. Kim EY, Park SA, Park BJ, Lee Y, Oh MM. Growth and antioxidant phenolic compounds in cherry tomato seedlings grown under monochromatic light-emitting diodes. Hortic Environ Biote. 2014;55:506-513.
8. Wu MC, Hou CY, Jiang CM, Wang YT, Wang CY, Chen HH, et al. A novel approach of LED light radiation improves the antioxidant activity of pea seedlings. Food Chem. 2007;101(4): 1753-1758.
9. Benedet JA, Umeda H, Shibamoto T. Antioxidant activity of flavonoids isolated from young green barley leaves toward biological lipid samples. J Agric Food Chem. 2007;55(14): 5499-5504.
10. Markham KR, Mitchell KA. The mis-identification of the major antioxidant flavonoids in young barley (*Hordeum vulgare*) leaves. Z Naturforsch C J Biosci. 2003;58(1-2):53-56.
11. Yu YM, Chang WC, Chang CT, Hsieh CL, Tsai CE. Effects of young barley leaf extract and antioxidative vitamins on LDL oxidation and free radical scavenging activities in type 2 diabetes. Diabetes Metab. 2002;28(2):107-114.
12. Yoshizawa M, Yokoyama K, Nakano Y, Nakamura H. Protective effects of barley and its hydrolysates on gastric stress ulcer in rats. Yakugaku Zasshi. 2004;124(8):571-575.
13. Mälkki Y. Trends in dietary fibre research and development. Acta Aliment. 2004;33(1):39-62.
14. Trepel F. Dietary fibre: More than a matter of dietetics: I. Compounds, properties, physiological effects. Wien Klin Wochenschr. 2004;116:465-476.
15. Jensen MK, Koh-Banerjee P, Hu FB, Franz M, Sampson L, Grønbæk M, et al. Intakes of whole grains, bran, and germ and the risk of coronary heart disease in men. Am J Clin Nutr. 2004 Dec 1;80(6):1492-1499.
16. Brennan CS. Dietary fibre, glycaemic response, and diabetes. Mol Nutr Food Res. 2005;49(6):560-570.
17. Brown L, Rosner B, Willett WW, Sacks FM. Cholesterol-lowering effects of dietary fiber: A meta-analysis. Am J Clin Nutr. 1999;69(1):30-42.
18. Hughes SA, Shewry PR, Gibson GR, McCleary BV, Rastall RA. In vitro fermentation of oat and barley derived β-glucans by human faecal microbiota. FEMS Microbiol Ecol. 2008;64(3):482-493.
19. Kontula P, von Wright A, Mattila-Sandholm T. Oat bran β-gluco- and xylo-oligosaccharides as fermentative substrates for lactic acid bacteria. Int J Food Microbiol. 1998;45(2):163-169.
20. Drzikova B, Dongowski G, Gebhardt E. Dietary fibre-rich oat-based products affect serum lipids, microbiota, formation of short-chain fatty acids and steroids in rats. Br J Nutr. 2005;94(6): 1012-1025.
21. Koga R, Meng T, Nakamura E, Miura C, Irino N, Devkota HP, et al. The effect of photo-irradiation on the growth and ingredient composition of young green barley (*Hordeum vulgare*). Agric Sci. 2013;4(4):1-10.
22. Chung NJ, Kim JY, Lee Y, Shin SH, Song JS, Shin SC, et al. Variations of saponarin content in young barley leaves illuminated with different Light-Emitting Diodes (LEDs). J Crop Sci Biotechnol. 2019;22:317-322.
23. Folin O, Denis W. A colorimetric method for the determination of phenols (and phenol derivatives) in urine. J Biol Chem. 1915;22(2):305-308.
24. Shimada K, Fujikawa K, Yahara K, Nakamura T. Antioxidative properties of xanthan on the autoxidation of soybean oil in cyclodextrin emulsion. J Agric Food Chem. 1992;40(6):945-948.
25. Bradley DW, Emery G, Maynard JE. Vitamin C in plasma: A comparative study of the vitamin stabilized with trichloroacetic acid or metaphosphoric acid and the effects of storage at -70°,-20°, 4°, and 25° on the stabilized vitamin. Clin Chim Acta. 1973;44(1): 47-52.
26. Kochetova GV, Avercheva OV, Bassarskaya EM, Kushunina MA, Zhigalova TV. Effects of red and blue LED light on the growth and photosynthesis of barley (*Hordeum vulgare* L.) seedlings. J Plant Growth Regul. 2023;42(3):1804-1820.
27. Beck E, Ziegler P. Biosynthesis and degradation of starch in higher plants. Annu Rev Plant Biol. 1989;40:95-117.
28. Fincher GB. Molecular and cellular biology associated with endosperm mobilization in germinating cereal grains. Annu Rev Plant Biol. 1989;40:305-346.
29. Lee NY, Lee MJ, Kim YK, Park JC, Park HK, Choi JS, et al. Effect of light emitting diode radiation on antioxidant activity of barley leaf. J Korean Soc Appl Biol Chem. 2010;53(6):685-690.
30. PauličkoVá I, EhrENbErgEroVá J, FIEdlEroVá V, Gabrovská D, Havlová P, Holasová M, et al. Evaluation of barley grass as a potential source of some nutritional substances. Czech J Food Sci. 2007;25(2):65-72.
31. Landete JM. Dietary intake of natural antioxidants: Vitamins and polyphenols. Crit Rev Food Sci Nutr. 2013;53(7):706-721.