Research article

Effects of LED Lighting and Media on Vitamin C and Phenol Content in Ethiopian Kale (*Brassica carinata*) Microgreens

RUTH MARU
School of Agriculture and Environmental Sciences,
Jomo Kenyatta University of Agriculture and Technology, Kenya

JOHN WESONGA
School of Agriculture and Environmental Sciences,
Jomo Kenyatta University of Agriculture and Technology, Kenya

DICKSON MAZIBUKO
Graduate School of Agro-Environmental Science, Tokyo University of Agriculture, Japan

SATOKO AKIYAMA
Faculty of Applied Bio-Science, Tokyo University of Agriculture, Japan

AYAKO SEKIYAMA
Faculty of Regional Environment Science, Tokyo University of Agriculture, Japan

SHOTARO KAWAKAMI
Faculty of Regional Environment Science, Tokyo University of Agriculture, Japan

SARVESH MASKEY
Faculty of Regional Environment Science, Tokyo University of Agriculture, Japan

AGNES KAVOO
School of Agriculture and Environmental Sciences,
Jomo Kenyatta University of Agriculture and Technology, Kenya

JOHNSTONE NEONDO
Institute for Biotechnology Research,
Jomo Kenyatta University of Agriculture and Technology, Kenya

HIROMU OKAZAWA*
Faculty of Regional Environment Science, Tokyo University of Agriculture, Japan
Email: h1okazaw@nodai.ac.jp

Received 30 December 2023 Accepted 17 December 2024 (*Corresponding Author)

Abstract Microgreens are innovative vegetable products related to their novelty and health-promoting benefits. However, growth media and light conditions affect microgreens' nutritional content, which may limit their production in rural community settings. *Brassica carinata* is an essential local Kenyan vegetable, but its production and full utilization are limited by its early maturity. The potential of using *B. carinata* as a microgreen would be an excellent alternative production technique to minimize its early maturity limitation. This study investigated the influence of white and blue light on Vitamin C and phenol content in *B. carinata* microgreens grown using sand and cocopeat. *B. carinata* microgreens were grown for 14 days in a growth chamber using plastic punnet containers filled with cocopeat and sand under white and blue light. The capillary wick watering technique was used for irrigation. Temperature and relative humidity were monitored and maintained at 26°C± 2, and 60%, respectively. The photoperiod and intensity of light were also maintained at 12 hr and 160 ± 2.5 µmol $m^{-2}s^{-1}$) respectively. After 14 days, microgreens were harvested and freeze-dried to analyze Vitamin C and phenol content. And. Data was subjected to ANOVA and was separated by Tukey's multiple comparison test. Results indicated that light had no significant effect on *B. carinata* microgreens phenol content. However, microgreens grown in locally available sand showed statistically higher amounts of phenol content than those grown using cocopeat. For vitamin C content, media and light had no significant effect. Our results show that sand medium can be used equally to produce microgreens with higher phenol content for *Brassica carinata.*

Keywords chronic diseases, growing media, LED lighting, micro greens, nutraceutical

INTRODUCTION

Microgreens, young and tender cotyledonary leafy greens of most vegetables, grains, and herbs (Michell et al., 2020) are also defined as immature vegetable greens harvested after cotyledonary leaves are developed (Zhang et al., 2021). Microgreens have recently gained public and research attention due to their potential as rich food resources. Microgreens have thus been called "functional foods" (Kalinová et al., 2023) with huge health benefits, especially in combating an array of non-communicable diseases (NCDs) and other chronic illnesses (Bhaswant et al., 2023; Zhang et al. 2021). In Sub-Saharan Africa, hidden hunger is prevalent, and little progress has been made over time (Ekholuenetale et al., 2020). Microgreens may provide a good alternative to alleviating undernutrition, especially in communities where poverty is prevalent.

Microgreen production media, a significant component of production costs, is a key determinant of yield and quality (Di Gioia et al., 2017). The high media cost has been noted as one limiting factor to upscaling microgreen production, prompting a search for alternatives (Thepsilvisut et al., 2023). Different low-cost media have been studied with varying results.

In addition to media, microgreen production is influenced by light, particularly light-emitting diodes (LEDs) treatments, which act as elicitors that trigger various biosynthetic pathways associated with different phytochemicals. The effects of light are further characterized by direction, duration of exposure, and intensity; all constituting a 'dose' (Artés-Hernández et al., 2022) for eliciting activation of singular pathways. Recent investigations into the effects of light on microgreen production demonstrate that altering the quality and type of spectra can enhance the accumulation of targeted phytochemicals (Kyriacou et al., 2016). As such, light optimization is crucial. Such optimization may require specificity to a desired phytochemical or other attributes. While many studies have defined the effects of light on microgreen phytochemicals, as reviewed by Artés-Hernández et al. (2022), Putri et al. (2022), Toscano et al. (2021), and Zhang et al. (2020) studied the effects and interactions between both light and media are rare.

OBJECTIVE

This study investigated the role of media and light on vitamin C and phenol content of *Brassica carinata*. Specifically, we compare vitamin C and phenol content of *B. carinata* microgreens grown in sand, an abundant resource in Africa, and cocopeat, under white and blue lighting regimes.

METHODOLOGY

Brassica carinata A. Braun, Ethiopian mustard, is an indigenous vegetable with origins in Ethiopia and parts of East Africa (Alemayehu and Becker, 2002), and which is commonly cultivated in Southern Africa, where it is utilized as a medicinal plant (Nakakaawa et al., 2023). Similar to most vegetable species in Southern Africa, *B. carinata* microgreens production dynamics are yet to be studied. To date, traceable studies on *B carinata* microgreens remain an assessment of the toxicity of *B carinata* microgreens (Nakakaawa et al., 2023) and the influence of salinity and light spectra of phytochemical production (Maina et al., 2021).

The experiment was conducted in a controlled environment in a locally fabricated walk-in growth chamber at the Tokyo University of Agriculture between April and October 2023. The chamber was divided into two compartments using a black opaque, fabric material to prevent light interference. Each compartment measured 1.0 m by 1.0 m. In each compartment, an LED light was placed 50 cm above the surface of the substrate. Ethiopian kale (*Brassica carinata*) seeds were sourced from a commercial vendor in Kenya.

B. carinata microgreens were sown and grown using two media, cocopeat and sand. and two LEDs in a factorial experiment. The light spectra were blue (450 nm) and cool white light in each compartment. The different LED lights were placed 50 cm above the growing media. An opaque black material was used to separate the different light spectra within the growth chamber. Each compartment contained two experimental units, cocopeat or sand, and one LED light in a split-plot design, with light as the main plot factor and media as the subplot factor. Four replications for light spectra and nine for the media were done. In each light treatment, a fixed light intensity of 160 ± 2.5 $\mu mol\ m^{-2}s^{-1}$ was maintained and was applied for a 12h/day photoperiod. Air temperature in the walk-in growth chamber was set and maintained at 26°C± 2, and humidity was maintained at 60% during the experiment. Temperature and relative humidity were monitored using a data logger. The growth substrate and the growing microgreens were irrigated using capillary wick technology throughout the growth period.

Phenol and Vitamin C are key food components that protect communities from chronic ailments including cardiovascular disease, cancer, diabetes, and other inflammatory conditions (Calderón-Pérez et al., 2021; Mutha et al., 2021). Vitamin C is a phytochemical with diverse health benefits, and microgreens are known to contain high levels of Vitamin C (Kathi et al., 2022; Kathi et al., 2023). Because of these advantages, Phenol and Vitamin C were included in this study. The estimation of total phenol was determined using the Folin-Ciocalteau method using Gallic acid as standard (Meas, et al., 2020). Vitamin C content was analyzed using a rapid reflectometric test, Reflectoquant ascorbic acid test, using an RQflex hand-held reflectometer (Merk, Darmstadt, Germany).

Statistical analysis was performed using R software, version (4.3.2). All data were subjected to ANOVA and differences among means were determined by Tukey's multiple comparison test at $p<$ 0.05.

RESULTS AND DISCUSSION

Effect of Media on Phenol and Vitamin C Content

The content of Phenol and Vitamin C in microgreens was compared between the two media (Fig. 1). Microgreens grown in the sand showed significantly higher amounts of phenol content compared to those grown using cocopeat media. *B. carinata* microgreens grown using sand had the highest vitamin C content (69 mg/100g D/W) compared to those grown using cocopeat (60 mg/100g D/W). Still, they were not statistically significant from those grown in cocopeat. Furthermore, phenol content in *B. carinata* grown using sand and cocopeat substrate was 1.01 mg/DW and 0.95mg/DW, respectively.

Research citing the use of sand in microgreen production are scarce. A study by Hoang and Vu (2022) that used sand-soil mix as one of the test media for *Brassica* microgreens found sand-soil mix as the least-performing media in all the attributes studied. Another study on broccoli microgreens (Sulistiya, 2021) reported that cocopeat outperformed sand media. The use of media where sand was was mixed with other media, did not assess media comparisons (Priti, et al., 2022).

Effect of LED on Phenol and Vitamin C Content

Vitamin C content in blue and white LEDs was approximately 67 mg/100g DW and 63 mg/100g D/W, respectively. The values for phenol in blue and white light were 1.03 mg/100g DW and 0.93 mg/100g DW respectively (Fig. 2). There was no significant difference in Vitamin C and phenol content between microgreens grown with blue and white lights. Other studies on assessments of light effects on growth and phytochemical accumulation, exhibited variability in Brassica microgreens (Kamal et al., 2020) grown under different red, blue, and green, lighting ratios. White light supplemented with red, blue, or ultraviolet-A, facilitated better accumulation of phytochemicals than white light alone (Gerovac, et al., 2016). Research shows that some light combinations promote growth while at the expense of phytochemical accumulation (Kamal, et al., 2020) and vice versa.

These results show that sand and white light can successfully produce *B. carinata* microgreens. Sand, a readily available resource, can eliminate the need for expensive peat and peat-based mixes as media. White light can be sourced from low-cost LED bulbs, replacing halogens and incandescent bulbs in most African countries (Enongene et al., 2017). Such a prospect entails a relatively cost-effective microgreen production in this region, enabling communities to tap into the diverse health benefits offered by microgreen consumption.

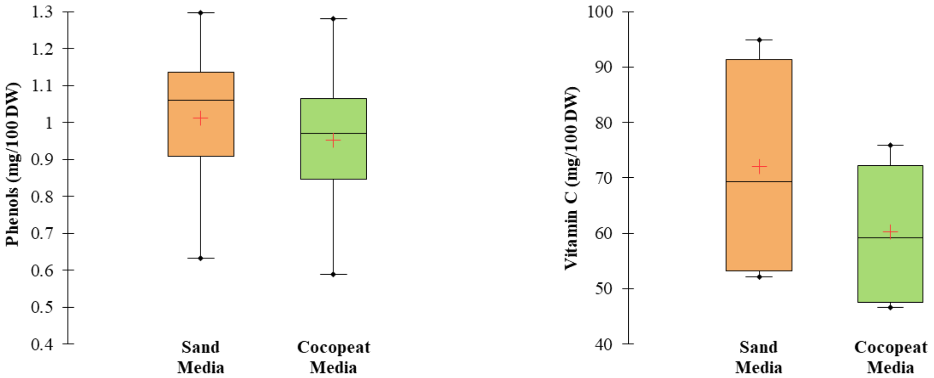

Fig.1 Phenol and vitamin C content for *B. carinata* microgreens grown in sand and cocopeat media

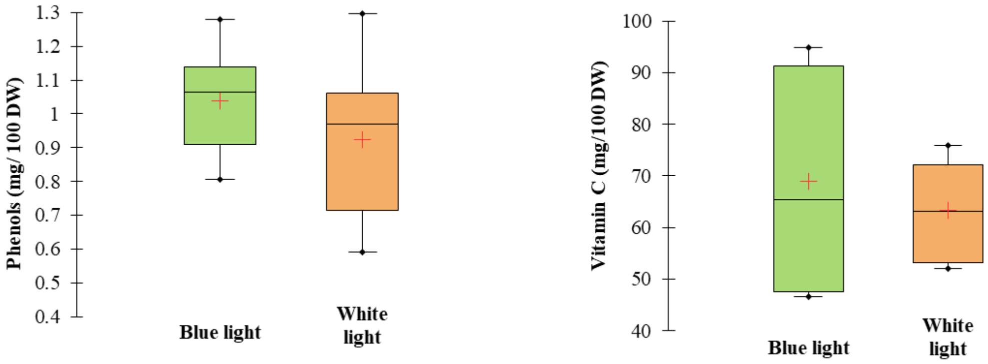

Fig. 2 Phenol and vitamin C content for *B. carinata* microgreens grown under blue and white LED lighting

CONCLUSION

The study aimed to assess the effects of media and lighting on the content of vitamin C and phenol in *B. carinata* microgreens. Results indicate that readily available sand and lights can be used as a production media targeting the two phytochemicals. These findings indicate an opportunity, particularly for Sub-Saharan Africa, where most chronic ailments that can benefit from the protective effects of microgreen phytochemicals are prevalent, to embrace the relatively cost-effective production using sand media. There remains a gap to investigate lighting optimization and media adjustments further to elucidate optimal doses for this species.

ACKNOWLEDGEMENTS

I would like to express my sincere gratitude to the Inter-University Exchange Programme of the Tokyo University of Agriculture and Jomo Kenyatta University of Agriculture and Technology (JKUAT). I would also like to thank JASSO for the scholarship and Tokyo University of Agriculture for hosting me throughout the project period. Part of the work was supported by the Africa- ai- Japan project under the JICA capacity building program at JKUAT to which I am indebted.

REFERENCES

Alemayehu, N. and Becker, H. 2002. Genotypic diversity and patterns of variation in a germplasm material of Ethiopian mustard (*Brassica carinata* A. Braun). Genetic Resources and Crop Evolution, 49, 573-582, Retrieved from DOI https://doi.org/10.1023/A:1021204412404

Artés-Hernández, F., Castillejo, N. and Martínez-Zamora, L. 2022. UV and visible spectrum LED lighting as abiotic elicitors of bioactive compounds in sprouts, microgreens, and baby leaves, A comprehensive review including their mode of action. Foods, 11 (3), 265, Retrieved from DOI https://doi.org/10.3390/foods 11030265

Bhaswant, M., Shanmugam, D.K., Miyazawa, T., Abe, C. and Miyazawa, T. 2023. Microgreens, A comprehensive review of bioactive molecules and health benefits. Molecules, 28 (2), 867, Retrieved from DOI https://doi.org/10.3390/molecules28020867

Calderón-Pérez, L., Llauradó, E., Companys, J., Pla-Pagà, L., Pedret, A., Rubió, L., Gosalbes, M.J., Yuste, S., Solà, R. and Valls, R.M. 2021. Interplay between dietary phenolic compound intake and the human gut microbiome in hypertension, A cross-sectional study. Food Chemistry, 344, 128567, Retrieved from DOI https://doi.org/10.1016/j.foodchem.2020.128567

Di Gioia, F., De Bellis, P., Mininni, C., Santamaria, P. and Serio, F. 2017. Physicochemical, agronomical and microbiological evaluation of alternative growing media for the production of rapini (*Brassica rapa* L.) microgreens. Journal of the Science of Food and Agriculture, 1212-1219, Retrieved from DOI https://doi.org/10.1002/jsfa.7852

Ekholuenetale, M., Tudeme, G., Onikan, A. and Ekholuenetale, C.E. 2020. Socioeconomic inequalities in hidden hunger, undernutrition, and overweight among under-five children in 35 sub-Saharan Africa countries. Journal of the Egyptian Public Health Association, 95, 9, Retrieved from DOI https://doi.org/10.1186/s42506-019-0034-5

Enongene, K.E., Murray, P., Holland, J. and Abanda, F.H. 2017. Energy savings and economic benefits of transition towards efficient lighting in residential buildings in Cameroon. Renewable and Sustainable Energy Reviews, 78, 731-742, Retrieved from DOI https://doi.org/10.1016/j.rser.2017.04.068

Gerovac, J.R., Craver, J.K., Boldt, J.K. and Lopez, R.G. 2016. Light intensity and quality from sole-source light-emitting diodes impact growth, morphology, and nutrient content of *Brassica* microgreens. HortScience, 51 (5), 497-503. Retrieved from DOI https://doi.org/10.21273/HORTSCI.51.5.497

Hoang, G.M. and Vu, T.T. 2022. Selection of suitable growing substrates and quality assessment of *Brassica* microgreens cultivated in greenhouse. Academia Journal of Biology, 44 (2), 133-142. Retrieved from DOI https://doi.org/10.15625/2615-9023/16833

Kalinová, J.P. and Nojeem, M.O. 2023. The quality of pasta with the addition of buckwheat microgreens or sprouts. Fagopyrum, 40 (2), 55-60. Retrieved from DOI https://doi.org/10.3986/fag0034

Kamal, K.Y., Khodaeiaminjan, M., El-Tantawy, A.A., Moneim, D.A., Salam, A.A., Ash-shormillesy, S.M.A.I., Attia, A., Ali, M.A.S., Herranz, R., El-Esawi, M.A., Nassrallahand, A.A. and Ramadan, M.F. 2020. Evaluation of growth and nutritional value of *Brassica* microgreens grown under red, blue and green LEDs combinations. Physiologia Plantarum, 169 (4), 625-638, Retrieved from DOI https://doi.org/10.1111/ppl. 13083

Kathi, S., Laza, H., Singh, S., Thompson, L., Li, W. and Simpson, C. 2022. Increasing vitamin C through agronomic biofortification of arugula microgreens. Scientific Reports, 12, 13093. Retrieved from DOI https://doi.org/10.1038/s41598-022-17030-4

Kathi, S., Laza, H., Singh, S., Thompson, L., Li, W. and Simpson, C. 2023. Vitamin C biofortification of broccoli microgreens and resulting effects on nutrient composition. Frontiers in Plant Science, 14, 1145992, Retrieved from DOI https://doi.org/10.3389/fpls.2023.1145992

Kyriacou, M.C., Rouphael, Y., Di Gioia, F., Kyratzis, A., Serio, F., Renna, M., De Pascale, S. and Santamaria, P. 2016. Micro-scale vegetable production and the rise of microgreens. Trends in food Science and Technology, 57 (A), 103-115, Retrieved from DOI https://doi.org/10.1016/j.tifs.2016.09.005

Maina, S., Ryu, D.H., Cho, J.Y., Jung, D.S., Park, J-E., Nho, C.W., Bakari, G., Misinzo, G., Jung, J.H., Yang, S-H. and Kim, H-Y. 2021. Exposure to salinity and light spectra regulates glucosinolates, phenolics, and antioxidant capacity of *Brassica carinata* L. microgreens. Antioxidants, 10 (8), 1183, Retrieved from DOI https://doi.org/10.3390/antiox10081183

Meas, S., Luengwilai, K. and Thongket, T. 2020. Enhancing growth and phytochemicals of two amaranth microgreens by LEDs light irradiation. Scientia Horticulturae, 265, 109204, Retrieved from DOI https://doi.org/10.1016/j.scienta.2020.109204

Michell, K.A., Isweiri, H., Newman, S.E., Bunning, M., Bellows, L.L., Dinges, M.M., Grabos, L.E., Rao, S., Foster, M.T., Heuberger, A.L., Prenni, J.E., Thompson, H.J., Uchanski, M.E., Weir, T.L. and Johnson, S.A. 2020. Microgreens, consumer sensory perception and acceptance of an emerging functional food crop. Journal of Food Science, 85 (4), 926-935, Retrieved from DOI https://doi.org/10.1111/1750-3841.15075

Mutha, R.E., Tatiya, A.U. and Surana, S.J. 2021. Flavonoids as natural phenolic compounds and their role in therapeutics, An overview. Future Journal of Pharmaceutical Sciences, 7, 25, Retrieved from DOI https://doi.org/10.1186/s43094-020-00161-8

Nakakaawa, L., Gbala, I.D., Cheseto, X., Bargul, J.L. and Wesonga, J.M. 2023. Oral acute, sub-acute toxicity and phytochemical profile of Brassica carinata A. Braun microgreens ethanolic extract in Wistar rats. Journal of Ethnopharmacology, 305, 116121, Retrieved from DOI https://doi.org/10.1016/j.jep.2022.116121

Priti, S.S., Kukreja, B., Mishra, G.P., Dikshit, H.K., Singh, A., Aski, M., Kumar, A., Taak, Y., Stobdan T, Das, S., Kumar, R.R., Yadava, D.K., Praveen, S., Kumar, S. and Nair R.M. 2022. Yield optimization, microbial load analysis, and sensory evaluation of mungbean (*Vigna radiata* L.), lentil (*Lens culinaris* subsp. *culinaris*), and Indian mustard (*Brassica juncea* L.) microgreens grown under greenhouse conditions. Plos One, 17 (5), e0268085. Retrieved from DOI https://doi.org/10.1371/journal.pone.0268085

Putri, E.A.D., Fajri, H.A.M., Iswari, F., Muhammad, F.A., Fauziah, R. and Budiarto, R. 2022. The impact of color of artificial LED lighting on microgreen, A review. Jurnal Kultivasi, 21 (2), 223-230, Retrieved from DOI http://dx.doi.org/10.24198/kultivasi.v21i2.39931

Sulistiya, S. 2021. Response to the growth and results of microgreens broccoli planted hydroponically with various planting media and addition of coconut water sources of nutrition and hormone. Journal Pertanian Agros, 23 (1), 217-229, Retrieved from URL https://lp3m.janabadra.ac.id/admin/assets/images/data_files_penelitian/1814_1339-2783-1-sm.pdf

Thepsilvisut, O., Sukree, N., Chutimanukul, P., Athinuwat, D., Chuaboon, W., Poomipan, P., Vachirayagorn, V., Pimpha, N., Chutimanukul, P. and Ehara, H. 2023. Efficacy of agricultural and food wastes as the growing media for sunflower and water spinach microgreens production. Horticulturae, 9 (8), 876, Retrieved from DOI https://doi.org/10.3390/horticulturae9080876

Toscano, S., Cavallaro, V., Ferrante, A., Romano, D. and Patané, C. 2021. Effects of different light spectra on final biomass production and nutritional quality of two microgreens. Plants, 10 (8), 1584, Retrieved from DOI https://doi.org/10.3390/plants10081584

Zhang, X., Bian, Z., Yuan, X., Chen, X. and Lu, C. 2020. A review on the effects of light-emitting diode (LED) light on the nutrients of sprouts and microgreens. Trends in Food Science and Technology, 99, 203-216, Retrieved from DOI https://doi.org/10.1016/j.tifs.2020.02.031

Zhang, Y., Xiao, Z., Ager, E., Kong, L. and Tan, L. 2021. Nutritional quality and health benefits of microgreens, A crop of modern agriculture. Journal of Future Foods, 1 (1), 58-66, Retrieved from DOI https://doi.org/10.1016/j.jfutfo.2021.07.001