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SCIENCE

Evaluation of various microgreen vegetables

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1. SUMMARY

As the world's population grows, more attention needs to be paid to producing foods that provide adequate nutritional value. Microgreens, which are becoming more and more popular today, can be considered such foods. Vegetables in the microgreen category become edible within 7 to 14 days when the cotyledons are fully developed and the first true leaves appear. Compared to adult plants, microgreens have a much higher nutritional value, they contain significant amounts of vitamins (ascorbic acid, tocopherol), minerals and phytonutrients. The amount of these bioactive substances is greatly influenced by environmental factors, including humidity, temperature and light intensity.

In our experiment, microgreens prepared species belonging to various plant families, such as *Brassicaceae* (mustard, radish), *Chenopodiaceae* (beetroot, Swiss chard) and *Lamiaceae* (basil), were evaluated based on the (Ca^{2+}+Na^{+})/(Mg^{2+}+K^{+}) ion ratio, yield, dry matter and vitamin C content.

Microgreens were allowed to develop up to a germination state of 90% protected from light in a germination chamber at a controlled temperature (24-25 °C) and humidity (65-70%). Following germination, after 2 to 3 days, the germination trays were placed in the experimental space of the greenhouse. General potting soil was used as the growing medium, and the seeds were of bio grade, suitable for microgreen cultivation in all cases. The crop was harvested 10 days after sowing, when plant height was 3 to 9 cm, depending on the species.

The highest dry matter content (~10%) was measured in species belonging to the family Chenopodiacea. Mustard contained an outstanding amount of vitamin C (22.66 mg/100 g). In addition, favorable biomass weights were found in the case of radish and mustard (2528 g/m^2; 1831 g/m^2), while the values for the other species were almost the same (~500 g/m^2).

The ion ratio of the human body varies between 2.5 and 4.0, with an optimal value of approximately 1.0. This relationship can be defined as the following ratio: (Ca^{2+}+Na^{+})/(Mg^{2+}+K^{+}).

The ratio is determined from the occurrence values of the elements expressed in mmol/l [27]. The consumption of vegetables is of paramount importance in establishing this ratio, as their ion ratio is mostly below 1.0. This health-improving effect was proven in the case of the microgreens examined by us, as a value of about 0.40 was obtained for the different species.

Overall, it can be stated that mustard showed the most favorable results for the parameters examined.

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2. Introduction

As the world's population grows, more attention needs to be paid to producing foods that provide adequate nutritional value and, in addition, have minimal environmental impact **[1, 2]**. Microgreens, which are becoming more and more popular today, can be considered such foods. These vegetable plants become edible within 7 to 14 days following germination, depending on the species, when the cotyledons are fully developed and the first true leaves appear **[3, 4, 5]**. Microgreens are therefore fundamentally different from sprouts **[6]** and baby leaf vegetable plants. The latter can be picked after a growing period of 20 to 40 days, in a mature, true leaf state. In contrast, sprouts are consumed before the emergence of the leaves, only the young shoots are consumed together with the seeds **[7]**. As consumer demand for healthy and so-called 'conveniance foods' grows, bagged and canned raw plants are becoming increasingly popular worldwide **[8]**. Recently, gastronomy developed a preference for them as well, as they play an important role in the offerings of exclusive restaurants with their special appearance. Their characteristic taste, which appears in a more pronounced way in the cotyledon state, can also be considered one of the favorable properties of microgreens **[9]**. They are used as new culinary ingredients to decorate various dishes, as side dishes or as raw materials for salad components **[5]**. Compared to the adult specimens of the same plants, microgreens have a much higher nutritional value, and they contain significant amounts of vitamins (ascorbic acid, tocopherol), minerals, carotenoids (β-carotene, lutein/zeaxanthin, violaxanthin) and phytonutrients **[4]**.

In the United States, the mineral content of thirty plant varieties produced as microgreens was evaluated in various food quality testing laboratories.

Ten species of the *Brassicaceae* family were evaluated for macro- and microelement content (Ca, Cu, Fe, K, Na Mg, Mn, P, Zn) using inductively coupled plasma optical emission spectrometry (ICP-OES). Based on their measurements, it was found that microgreens are good sources of both macroelements (K and Ca) microelements (Fe and Zn) **[10]**. In further experiments, the mineral and nitrate contents of lettuces produced as microgreens and mature specimens of the same plant species were compared. The results showed that most of the minerals (Ca, Mg, Fe, Mn, Zn, Se and Mo) were present in higher amounts in the microgreens. Additionally, they had a lower nitrate content than the adult specimens, and could therefore be considered an excellent and safe source of minerals in human nutrition **[11]**. The amount of these bioactive substances is greatly influenced by environmental factors, including temperature, humidity and light intensity. For this reason, the production conditions of microgreens fundamentally determine the quality and nutritional value of the product **[12]**. Furthermore, due to the short growing period, ensuring the best possible environmental conditions is of paramount importance **[13]**. Increased attention should be paid to the selection of appropriate species and production conditions (growing medium, optimal plant density, light intensity, temperature, humidity), in order to be able to protect them against possible fungal or bacterial infections **[14]**. Although in the case of these herbaceous, cotyledonous plants, the main sources of infection most often are pathogens on the surface of the seeds. Due to the risk of infection, it is advisable to disinfect seeds of poor quality, e.g., using a 20,000 mg/l calcium hypochlorite $Ca(OCl)_2$ solution **[15].** Furthermore, in the above-mentioned three plant product categories (sprout, microgreen, baby leaf), there are strict production and marketing standards only for sprouts. The reason for this is that the consumption of food sprouts produced under inadequate hygiene conditions poses a high food safety (microbiological) risk **[16]**.

Microgreens are usually produced in a closed environment, such as a greenhouse, foil tent or germination chamber, under natural or artificial lighting **[6, 17]**. Closed growing environments can also limit the access of insects and other organisms, thereby minimizing the possibility of infection **[8]**.

Microgreens typically produced include species of the *Brassicaceae* family, such as cabbage, radish, cauliflower, broccoli, and many herbs and spices, such as mustard and garden cress **[18]**. For species belonging to this plant family, it is well known that they are rich sources of glucosinolates and other phytochemicals with antioxidant effects, so that by consuming them, valuable, biologically active substances can enter the human body **[19, 20]**.

3. Materials and methods

Our experiments were set up in the CSK 7,56/2018 germination chamber found in the demonstration garden of the University of Debrecen AKIT DTTI Arboretum. During the growing, species belonging to the different plant families of *Brassicaceae* (mustard, radish), *Chenopodiaceae* (Swiss chard, beetroot) and *Lamiaceae* (basil) were evaluated. Sowing was performed on February 20, 2020, in 58 x 29 cm propagation trays by covering the seeds with a thin layer of soil, and then placing the trays in the germination chamber. The microgreens were kept under light-free conditions with controlled temperature (24-25 °C) and humidity (65-70%) until germination was 90% complete. General potting soil was used as the growing medium. In all cases, the seeds were organically certified, suitable for growing seedlings and microgreens. After a light-free period of 2 to 3 days, the trays were placed in the separate experimental space of the greenhouse on the demonstration garden. Temperature, humidity and irradiance values were measured at 2 typical times of the assimilation period, in the morning (between 8 and 9 a.m.) and in the afternoon (between 1 and 2 p.m.), checking the lower and higher temperature intervals of the active photoperiodic section. It can be clearly seen that during this period the temperature was in the range of 17.75±1.28 °C, while in the afternoon it was in the range of 22.13±3.48 °C (***Figure 1***).

Humidity varied between 60±7.8% in the morning and 51±13.2% in the afternoon. Irradiance was in the range of 27±17.3 W/m^2 in the morning and in the range of 96±65.5 W/m^2 in the afternoon. Light intensity varied between 2353±1206 lx in the morning and 2277±606 lx in the afternoon (***Figure 2***).

Harvesting of the crop was carried out after 10 days, on March 1, 2020, by cutting the 3 to 9 cm tall plants without the roots along the soil surface.

Data were compared using the analysis of variance (ANOVA) method. Among the post-hoc tests, the difference between the data at a significance level of 5% (p=0,05) was determined by Tukey's test. Analyses were performed using the IBM SPSS software (version 25).

3.1. Laboratory measurements

The aim of our experiments was to evaluate the nutritional parameters (ion ratio, dry matter and vitamin C content) and yield of species belonging to different plant families (*Brassicaceae* - mustard, radish), *Chenopodiaceae* – Swiss chard, beetroot, *Lamiaceae* - basil) when produced as microgreens.

In the course of our work, the following measurements were performed:

- Determination of biomass weight (g/m^2) at harvest for each species;
- Determination of total dry matter content (%) – according to Chapter 2 of standard MSZ-08-1783-1:1983;
- Determination of ascorbic acid content – according to Chapter 2 of standard MSZ ISO 6557-2:1991;
- Determination of mineral content (Ca, K, Mg, Na) with ICP-AES. Sample preparation was carried out by dry ashing according to standard MSZ-08-1783:1983. Measurement results were used to calculate the ion ratio.

4. Analytical results

4.1. Dry matter content

Within the dry matter content, two components are distinguished, organic and inorganic substances. The dry matter content determines the raw shelf life of plants, which is an important property for microgreens. The dry matter content of the samples evaluated by us ranged from 4.78% to 10.02% (***Table 1***). The highest values (~10%) were measured in samples prepared from beetroot and Swiss chard. The lowest dry matter (4.78%) was found in radish. Other researchers have measured dry matter contents of 7.73% in basil, 7.8% in radish, and 4.6 and 5.6% in beetroot and mustard, respectively, under different conditions in unheated foil tents (May and July) **[4]**.

4.2. Vitamin C content

Ascorbic acid (vitamin C) is one of the water-soluble vitamins, so it is not stored in the human body, therefore, it must be constantly replenished. It is a well known fact that the recommended daily intake values are controversial for several nutrients. According to the RDA (Recommended Dietary Allowance), the recommended daily intake of vitamin C for the general population is 90 mg/day for adult men and 75 mg/day for adult women **[21]**. The vitamin C content of the vegetable species evaluated by us ranged from 2.44 to 22.66 mg/100 g (***Table 2***). The latter value was measured in mustard, and it can cover 25 to 30% of the recommended daily intake. When using hydroponic cultivation technology, other researchers have measured values of 30.67 mg/100 g in mustard and 45.43 mg/100 g in radish **[22]**.

4.3. Ion ratios of the different microgreens

The optimal ion ratio of the human body can be defined by the following relation: $(Ca^{2+}+Na^{+})/\ Mg^{2+}+K^{+}) \approx 1.0$, where the ratio of mineral elements is expressed in mmol/l values. In the human body, this value usually varies between 2.5 and 4.0 **[27]**, and this ion ration can be favorably influenced by the consumption of vegetables **[22]**.

Such a health-improving effect of the microgreens examined by us could be proven, as in most cases the calculated ion ratios were around 0.40 for the different vegetable species (***Table 3***). The highest ion ratio was obtained for basil (0.62). Thus, it can be proven that the consumption of microgreens can lower the ion ratio of the human body, making it approach the optimal value (1.0).

4.4. Yields of microgreens produced from different plant species

When producing microgreens, typically lower yields are to be expected. ***Table 4*** shows the biomass weight measured for each species. In our experiment, outstanding yields were measured for radishes and mustard with typically larger leaves, specifically 2,528 g/m^2 for radishes and 1,831 g/m^2 for mustard. This value was lower for the Swiss chard, beetroot and basil species. Other researchers achieved yields of ca. 2,000 g/m^2 for Swiss chard and ca. 1,000 g/m^2 for basil using hydroponic cultivation **[24]**. Murphy et al. used different sowing (seed) densities in the production of microgreens from beetroot, which was harvested after 15 days. It was found that although higher seed densities resulted in higher yields, but a fourfold increase in the initial seed amount of 50 g/m^2 resulted in a yield increase per square meter of only 60% **[25]**.

5. Evaluation and discussion of the results, conclusions

Microgreens are young, tender herbaceous plants that wither in a relatively short time, so it is advisable to grow species that have a more favorable (lower) water content.

In our experiment, when grown on peat, the highest water content was measured in radish (95.2%), while the lowest was measured in Swiss chard (89.8%).

For a balanced diet, it is important to have the right amount of vitamin C. As vitamin C is not stored in the human body, it must be constantly replenished. Of the vegetable species examined, a favorable vitamin C content was measured in the case of mustard (22.66 mg/100 g), which can cover 25 to 30% of the recommended daily intake.

All the vegetable species evaluated by us proved to be favorable in maintaining the optimal ion ratio of the human body.

In the literature available to us, relatively little data was found on the yield of the different species, while using different cultivation methods. From an economic point of view, it would be important to know the yields that can be expected, because of the relatively high seed demand (10-40 thousand seeds/m^2) **[26]**.

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7. References

[1] Weber, F. C. (2017): Microgreen Farming and Nutrition: A Discovery-Based Laboratory Module to Cultivate Biological and Information Literacy in Undergraduates. The American Biology Teacher 79 (5): 375-386

[2] McClung, C. R. (2014): Making Hunger Yield. Plant Science, AAAS. 344 (6185): 699–700.

[3] Bhatt, P., Sharma, S. (2018): Microgreens: A Nutrient Rich Crop that can Diversify Food System. Internatioanl Journal of Pure & Applied Bioscience. 6 (2): 182-186.

[4] Xiao, Z., Lester, G.E., Luo, Y., Wang, Q. (2012): Assessment of Vitamin and Carotenoid Concentrations of Emerging Food Products: Edible Microgreens. Journal of Agricultural and Food Chemistry 60 (31): 7644 –7651.

[5] Nivedha, V., Lakshmy, P. S. (2018): Comparative study of microgreens with mature greens incorporated ready-to-eat chutney powders. International Journal of Food Science and Nutrition. 3 (6): 171-175

[6] Kaiser, C., M. Ernst. (2018): *Microgreens*. CCD-CP-104. Lexington, KY: Center for Crop Diversification, University of Kentucky College of Agriculture, Food and Environment.

[7] Di Gioia, F., Renna, M., & Santamaria, P. (2017): Sprouts, Microgreens and "Baby leaf" Vegetables. In *Minimally Processed Refrigerated Fruits and Vegetables.* Springer, Boston, MA. **403**-432.

[8] Xiao, Z., Nou, X., Luo, Y., Wang, Q. (2014): Comparison of the growth of Escherichia coli O157: H7 and O104: H4 during sprouting and microgreen production from contaminated radish seeds. Food Microbiology, 44, 60–63.

[9] Takácsné Hájos M., Kovácsné Madar Á. (2020): Változó fogyasztási szokások – mikrozöldésegek. Agrofórum Iránymutató a mezőgazdaságban. 1 (31): 52–54.

[10] Xiao, Z., Codling, E. E., Luo, Y., Nou, X., Lester, G. E., & Wang, Q. (2016): Microgreens of Brassicaceae: Mineral composition and content of 30 varieties. Journal of Food Composition and Analysis, 49: 87–93.

[11] Pinto, E., Almeida, A. A., Aguiar, A. A., Ferreira, I. M. P. L. V. O. (2015): Comparison between the mineral profile and nitrate content of microgreens and mature lettuces. Journal of Food Composition and Analysis, 37, 38–43.

[12] Nolan, D. A. (2018): Effects of Seed Density and Other Factors on the Yield of Microgreens Grown Hydroponically on Burlap. Virginia Tech. 39.

[13] Delian, E., Chira, A., Badulescu L., Chira, L. (2015): Insights into microgreens physiology. Scientific Papers. Series B, Horticulture. LIX, 447-454.

[14] Kyriacou, M. C., Rouphael, Y., Di Gioia, F., Kyratzis, A., Serio, F., Renna, M., De Pascale, S., Santamaria, P. (2016): Micro-scale vegetable production and the rise of microgreens. Trends in Food Science & Technology. 57. 103–115.

[15] National Advisory Committee on Microbiological Criteria for Foods (1999): Microbiological safety evaluations and recommendations on sprouted seeds. International Journal of Food Microbiolgy 52 (3): 123-153

[16] Treadwell, D. D., Hochmuth, R., Landrum, L., Laughlin, W. (2010): Microgreens: A New Specialty Crop. University of Florida IFAS Extension, HS1164. 1-3.

[17] Stoleru, T., Ioniță, A., Zamfirache, M. M. (2016): Microgreens - a new food product with great expectations. Romanian journal of biology, Plant Biology. 61 (1-2): 7-16.

[18] Paradiso, V. M., Castellino, M., Renna, M., Gattullo, C. E., Calasso, M., Terzano, R., Allegretta, I., Leoni, B., Caponio, F., Santamaria, P. (2018): Nutritional characterization and shelf-life of packaged microgreens. Food & Function. 9 (11): 5629–5640.

[19] Kopsell, D. A., Sams, C. E. (2013): Increases in Shoot Tissue Pigments, Glucosinolates, and Mineral Elements in Sprouting Broccoli after Exposure to Short-duration Blue Light from Light Emitting Diodes. Journal of the American Society for Horticultural Science. 138. (1): 31-37.

[20] Samuolienė, G., Brazaitytė, A., Viršilė, A., Miliauskienė, J., Vaštakaitė-Kairienė, V., Duchovskis, P. (2019): Nutrient Levels in Brassicaceae Microgreens Increase Under Tailored Light-Emitting Diode Spectra. Frontiers in Plant Science, 10. 1475

[21] Institute of Medicine (2000): Dietary Reference Intakes for Vitamin C, Vitamin E, Selenium, and Carotenoids. Washington, DC: The National Academies. 528.

[22] De la Fuente, B., López-García, G., Mañez, V., Alegría, A., Barberá, R., Cilla, A. (2019): Evaluation of the Bioaccessibility of Antioxidant Bioactive Compounds and Minerals of Four Genotypes of Brassicaceae Microgreens. Foods. 8 (7): 250.

[23] Takácsné Hájos M., Kiss A. S., Kastori, R., Ravel, J., Stefanovits-Bányai É., Sekulic, P. (2005): Evolution of protein and mineral element contents in green pea varieties as affected by Mg-sulphate leaf fertilisation. TSF Tudományos Közlemények. 5 (2): 59-71.

[24] Bulgari, R., Baldi, A., Ferrante, A., Lenzi, A. (2016): Yield and quality of basil, Swiss chard, and rocket microgreens grown in a hydroponic system. New Zealand Journal of Crop and Horticultural Science. 45 (2): 119–129.

[25] Murphy, C. J., Llort, K. F., Pill, W. G. (2010): Factors Affecting the Growth of Microgreen Table Beet. International Journal of Vegetable Science, 16 (3): 253–266.

[26] Di Gioia, F., Santamaria, P. (2015): Microgreens. Bari: Eco-logica editore 116.

[27] Balla Á., Kiss, A. S. (1996): Magnézium a biológiában. Magnézium a gyermekgyógyászatban. *Pro-Print Kiadó,* Csíkszereda 449 p.

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