

# Basil and Lettuce Microgreens Production in Low-Cost Hydroponic Installations, under Operational and Semi-Controlled Conditions

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## RESEARCH ARTICLE

### Abstract

Microgreens produced hydroponically at home, represent a solution of the future due to the advantages it has. The purpose of this research is to analyze certain elements relevant for the microgreens crop (basil and lettuce), simultaneously, in identical low-cost hydroponic platforms, in operational (common spaces) and semi-controlled conditions (plant growing tent). The obtained results were analyzed, both from the point of view of the growing conditions and the influence of light regimes, as well as the development of seedlings, production (fresh weight and dry weight) and quality (total phenolic) of the microgreens. In operational environmental conditions there is a higher variation of the environmental parameters, compared to the semi-controlled environment, but the oscillation of natural light seems to have a better influence on the development of microgreens. Internode length in operational environmental conditions varies for basil between 25.2-35.2 mm, for lettuce between 31.9-32.8 mm, and in semi-controlled environmental conditions, it varies for basil between 25.2-32.1 mm and for lettuce between 30.2-31.9 mm. In semi-controlled conditions, the average fresh weight production achieved by the two species (and four varieties) is 734.73 g m<sup>-2</sup> for basil, 809.56 g m<sup>-2</sup> for lettuce, and represents 823.74 g m<sup>-2</sup> for basil, 777.61 g m<sup>-2</sup> for lettuce in operational environmental conditions. The variability of the data recorded for total phenols is very high. Lettuce contains 98-107.33 mg kg<sup>-1</sup>, and basil 107.67-186.33 mg kg<sup>-1</sup> total phenols. Feedback from these tests will be used for final validation of the low-cost hydroponic platform components.

**Keywords:** functional food; microgreens; hydroponic; operational and semi-controlled conditions

## INTRODUCTION

Microgreens, considered the functional food of the 21st Century (Choe and Wang, 2018) are seen as important crops in the agriculture of the future (Bhaswant et al., 2023). They are mainly produced hydroponically or semi-hydroponically with very short growth cycles (usually 7-21 days after germination) having numerous advantages (Galieni et al., 2020; Bulgari et al., 2021; Ebert, 2022; Bhaswant et al., 2023). Hydroponic cultivation has emerged as a solution of the future because it does not require arable land, reduces the use of clean water (Velazquez-Gonzalez et al., 2022; Elmulthum et al., 2023; Sela Saldinger et al., 2023) and can be used in any urban environment (Naqvi et al., 2022; Pomoni et al., 2023). Microgreens are eaten as young plants, in a seedling stage (Martinez-Ispizua et al., 2022), having a high density of nutrients (much higher than the mature plant per unit weight)


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(Fraszczak et al., 2022), have a short production cycle (Mezeyova et al., 2022) and need little space to grow (Rouphael et al., 2021; Kelly et al., 2022). Microgreens can be planted from vegetables, cereals, technical plants or herbs consumed in the stage of tender cotyledons (Pignata et al., 2017; Ebert, 2022; Fraszczak et al., 2022), and the first pairs of leaves are more or less developed (Martinez-Ispizua et al., 2022). At harvest, the height of the seedling varies from 2.5 to 8 cm depending on the species (Lenzi et al., 2019; Mezeyova et al., 2022). Growth is achieved in conditions of intense light, low humidity and good ventilation (Amitrano et al., 2020; Hemathilake et al., 2022). The aroma, appearance and nutritional properties are completely different, improved, compared to the sprouts or the mature plant (Keutgen et al., 2021; Puccinelli et al., 2022). Other advantages are related to the reduced consumption of energy, water and agricultural land, reduced pollution of the environment, and does not require pesticides and fertilizers (Ciuta et al., 2020; Van Gerrewey et al., 2022). This type of culture also lends itself to intensive automation, which is also important (Galieni et al., 2020; Fussy et al., 2022). By producing microgreens in your own kitchen, they can be consumed without heat processing (Di Gioia et al., 2016; Hong and Gruda, 2020), which preserves active substances that would otherwise be lost (such as enzymes) (Buturi et al., 2021; Zagorska et al., 2022). Microgreens play an important role in cures and complex treatments, which include natural diets, developed by researchers from Europe and the USA (Sreenivasa et al., 2019; Renna et al., 2020).

One of the major limitations to the expansion of the consumption of microgreens is the rapid deterioration of their quality, which occurs immediately after harvesting (Turner et al., 2020), thus limiting their commercialization (Mlinaric et al., 2023). From this point of view, the development of low-cost cultivation platforms represents an optimal solution in the future, microgreens being very suitable for hydroponic production on different substrates, indoors, representing a sustainable alternative of urban agriculture, within the reach of any family (Ampim et al., 2022; Sela Saldinger et al., 2023).

In this context, the GoHydro project (<https://www.gohydro.org/>; accessed on 19 February 2023) aims to develop a platform based on artificial intelligence, cost-effective, capable of monitoring the phytosanitary status of crops and the nutrient content of micro-plants grown hydroponically, in order to optimize the cultivation process and allow obtaining the best possible products. The research presented in this paper is carried out within this research project.

The purpose of this research is to analyze certain elements relevant for the microgreens crop: basil and lettuce, grown under hydroponic systems, established by the trial protocol of the GoHydro project presented by Moraru et al., 2022, such as: air temperature, water temperature, humidity, pH, electrical conductivity and the influence of light regimes (semi-controlled and environments settings). The researches were carried out simultaneously, in identical low-cost hydroponic platforms, in operational conditions (common spaces) and semi-controlled (plant growing tent). The obtained results were analyzed, both from the point of view of the growing conditions, as well as the production and quality (total phenolic) of the microgreens. The goal of all these determinations is to contribute to the development of knowledge concerning the challenges of microgreens crops in a hydroponic environment which will push forward discussions and practice in this segment of research and industry.

## MATERIALS AND METHODS

In order to meet the requirements of the project so that the datasets generated in each trial platform are comparable and combinable into a single, common dataset, the following principles for the elaboration of the hydroponic microgreen production protocol were established (Moraru et al., 2022):

- In semi-controlled settings – setting the optimal ranges of environmental parameters between the limits of favorability for each species, as seen in the literature review for microgreens to highlight the effects of the GoHydro platform; The optimal conditions are published by Rusu et al., 2021a for basil and by Rusu et al., 2021b for lettuce.

- In operational environments settings – monitoring, but not controlling, environmental conditions (which will also be influenced by external environmental conditions); operational environments include office and living spaces for everyday use.

- It is also important to use the same type of substrate for seed germination and growth, and to use the same variety; common protocols has be set for data collection, data formatting, and collection intervals; data collection has be done using three repetitions.

During the experiment, they were built two GoHydro platforms, identical, to be able to do experiments in parallel, both in semi-controlled conditions and in operational conditions. The construction stages of the GoHydro platform are presented in Figure 1. For semi-controlled conditions we used the grow tent Eden Grow XL Growbox Growtent homegrowing 2.4 x 1.2 x 2.0 m Indoor.

GoHydro hydroponic platforms were built between February and October 2022, with the following systemically grouped components: Vertical hydroponic structure with three layers for growing microgreens; System for irrigation and recirculation; System for lighting and automation. Technical characteristics of the systems included in the GoHydro hydroponic platform are presented in Table 1.



**Figure 1:** Construction of individual platforms (a) and Eden Grow tent (b)

**Table 1.** Vertical hydroponic structure with three layers for growing microgreens

No	Structure	Characteristics
1	Number of modules	2
2	Type of structure/assembly	Demountable
3	Thickness of the system structure, mm	20
4	Hydroponic system length, mm	800
5	Width hydroponic system, mm	450
6	Hydroponic system height, mm	1400
7	No. of floors of culture/module	3
8	Height to first floor, mm	400
9	Distance between floors, mm	300
10	No. of culture trays/module	3
11	Culture tray length, mm	670
12	Culture tray width, mm	320
13	Culture tray height, mm	30
14	LED bulbs 6500K - 18W	6

The experiment was carried out with 2 species, basil (*Germaline Basilic Bio* and *Basil Grand Vert*) and lettuce (*Paris White*, *Little Gem*, and *Lolla Rossa*) varieties:

**1. For the basil:** tray size (3): 64 cm x 31 cm = 1984 cm<sup>2</sup>; each one with two varieties = 992 cm<sup>2</sup>: *Germaline Basilic Bio* (Italiano classico) = 3.84 g tray<sup>-1</sup> (992 cm<sup>2</sup>); *Basil Grand Vert* = 3.78 g tray<sup>-1</sup> (992 cm<sup>2</sup>); sowing date: 4/11/2022, 48 hours in the dark for germination; substrate - hemp.

**2. For the lettuce:** tray size (3): 64 cm x 31 cm = 1984 cm<sup>2</sup>; each one with three varieties = 661 cm<sup>2</sup>: *Paris White* = 4.18 g tray<sup>-1</sup> (661 cm<sup>2</sup>); *Little Gem* = 3.87 g tray<sup>-1</sup> (661 cm<sup>2</sup>); *Lolla Rossa* = 3.98 g tray<sup>-1</sup> (661 cm<sup>2</sup>); sowing date: 5/12/2022, 48 hours in the dark for germination; substrate - hemp.

The following devices were used for the measurements: Laserliner 082.130A LuxTest-Master Illumination Meter (Umarex GmbH & Co, Arnsberg, Germany); wtw pH 315i - Digital pH Meter (WTW Laboratory, London, United Kingdom); Milwaukee Pro Portable Meters (Milwaukee Instruments, Rocky Mount, United States).

Drinking water used for irrigation has the following characteristics: pH = 6.81, electrical conductivity (EC) = 0.261 dS m<sup>-1</sup>, total dissolved solids (TDS) = 389 ppm, nitrites - NO<sub>2</sub> = 0 mg L<sup>-1</sup>, nitrates - NO<sub>3</sub> = 0 mg L<sup>-1</sup>, and hardness = 14 °d. No nutrients were used.

The density was determined with the Microgreens Seed Density Calculator (Created by Francesco Di Gioia, available online; accessed on 4 November 2022).



Data collection was carried out according to a trial protocol for evaluating platforms for growing microgreens in hydroponic conditions (Moraru et al., 2022). The variables determined and the methodology used were as follows:

Determining the health state of plants, it was realized through the continuous monitoring of all the symptoms that appear. The health state of plants shall be noted in ascending order with grades from 1 to 9 (FAO grades), with the maximum grade corresponding to a perfect health state.

Measurements on the morphology of plants is performed in the juvenile vegetative phase before harvesting the microgreens. The surface of the leaves (leaves area) it was determined with a planimeter on 10 plants per tray. Ten representative plants for each tray of the platform it was harvested on the diagonals of the tray.

Fresh biomass yield. All the microgreens within each tray it was cut right above the substrate level (cutting them at the base, excluding the substrate) and collected to determine Fresh Weight (FW,  $\text{kg m}^{-2}$ ).

Dry Weight (DW,  $\text{g m}^{-2}$ ) it was measured on an analytical balance following lyophilization until a constant weight was reached. Each sample it was dried in an oven at  $70\text{ }^{\circ}\text{C}$  during 3 days until constant weight was reached.

Bioactive compound and total polyphenols were analyzed by HPLC methodology.

Experimental data were processed by statistical variant analysis with ANOVA PoliFact Soft, and the limit differences for p-values were established at 0.05, 0.01, and 0.001. For the Duncan test the limit differences for p-values were established at 0.05. For the statistical processing, the production was transformed into  $\text{g m}^{-2}$  for fresh weight and dry weight, and into  $\text{mg kg}^{-1}$  for total phenols. The statistical processing was carried out on each species separately, the average per species being taken as a control.

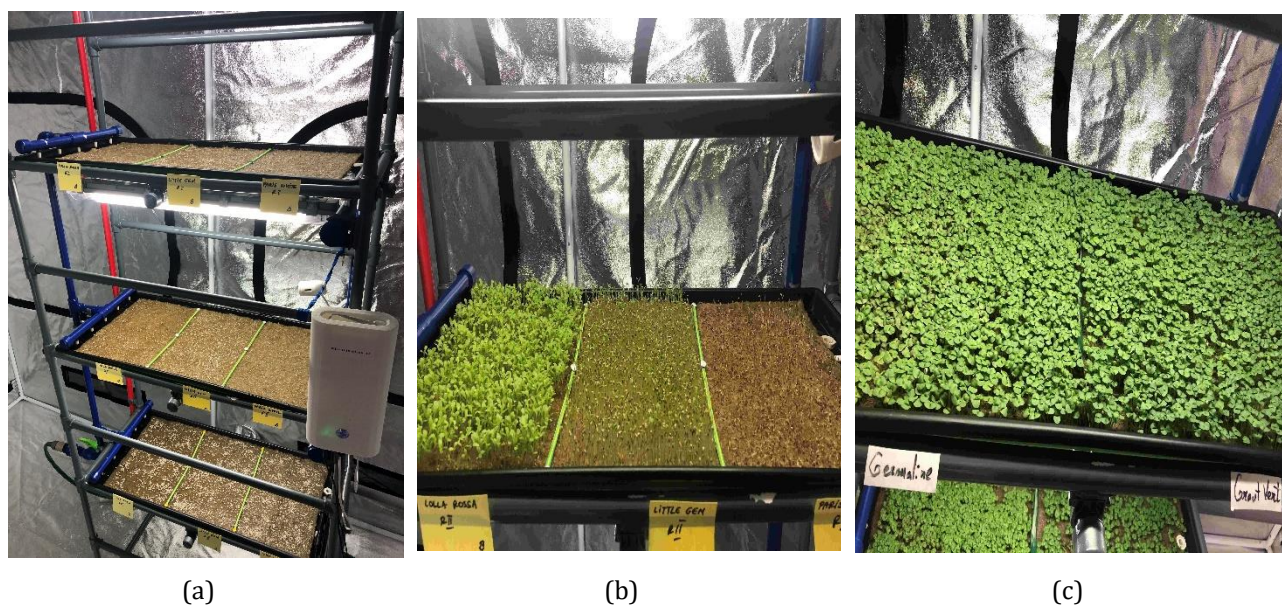
## RESULTS

### Parameters collected related to the growth environment in semi-controlled settings

The growing conditions of the two species of microgreens (basil and lettuce) are specific to semi-controlled conditions, using the Eden Grow plant growing tent. Thus, a temperature was kept in the atmosphere with a maximum oscillation of  $3\text{-}4\text{ }^{\circ}\text{C}$ , an atmospheric humidity oscillation of  $8\text{-}10\%$ , and respectively a constant amount of light (of  $1217\text{ }\mu\text{mol m}^{-2}\text{s}^{-1}$ ).

The irrigation water had a constant temperature of  $19\text{-}21\text{ }^{\circ}\text{C}$ , a pH of  $6.8\text{-}7$ , an electrical conductivity between  $1.4\text{-}1.5\text{ dS m}^{-1}$ , and respectively a quantity of  $6.4\text{-}6.5\text{ mg L}^{-1}$  dissolved oxygen.

The two experiments are presented in Figure 2. Parameters related to microgreen growth in semi-controlled settings, production and quality in the basil experiment is presented in Table 2, and for lettuce in Table 3. It can be noted, first of all, that in the case of lettuce, the Paris White variety, it did not germinate and as a result the production of microgreens of this variety was compromised. This result shows that the suitability of the variety used and the quality of the seeds is very important in the success of microgreens crops.



**Figure 2.** Experiment with seed of the lettuce (a), lettuce after 7 days from sowing (b), basil after 18 days from sowing (c) in semi-controlled conditions

**Table 2.** Parameters related to microgreen growth, production, and quality in the basil experiment in semi-controlled conditions

Parameter	Unit of measurement	Day 18/21.11.2022	Day 18/21.11.2022
<b>Determining the health state of plants</b>		<i>Germaline</i>	<i>Grand Vert</i>
Intensity of the disease attack	Note 1-9	1	1
<b>Measurements on the morphology of plants*</b>		r1/r2/r3	r1/r2/r3
Stem length	mm	31.2/32.1/32.1	25.2/25.4/25.2
Internode length	mm	31.2/32.1/32.1	25.2/25.4/25.2
Number of leaves	no	2/2/2	2/2/2
Number of plants	no cm <sup>-2</sup>	10.2/10.6/9.9	8.7/9.1/8.9
Leaves area (10 plants per tray)	cm <sup>2</sup>	0.52/0.54/0.52	0.49/0.52/0.51
<b>Determining fresh biomass yield and dry matter content</b>			
Fresh weight	g tray (992 cm <sup>2</sup> )	78.45/76.84/77.89	68.12/68.45/67.56
Dry weight	g tray (992 cm <sup>2</sup> )	7.79/7.69/7.81	6.78/6.81/6.78
Total phenols	mg kg <sup>-1</sup>	115/169/139	172/114/273

Note: \*r1- top tray; r2 - middle tray; r3 - bottom tray

**Table 3.** Parameters related to microgreen growth, production, and quality in the lettuce experiment in semi-controlled conditions

Parameter	Unit of measurement	Day 15 /19.12.2022	Day 15 /19.12.2022	Day 15 /19.12.2022
<b>Determining the health state of plants</b>		<i>Paris White</i>	<i>Little Gem</i>	<i>Lolla Rossa</i>
Intensity of the disease attack	Note 1-9	3	1	1
<b>Measurements on the morphology of plants*</b>		r1/r2/r3	r1/r2/r3	r1/r2/r3
Stem length	mm	0/0/0	30.4/31.1/30.2	31.4/31.8/31.9
Internode length	mm	0/0/0	30.4/31.1/30.2	31.4/31.8/31.9
Number of leaves	no	0/0/0	2.5/2.4/2.6	2.3/2.1/2.2
Number of plants	no cm <sup>-2</sup>	0/0/0	10.1/10.4/10.2	9.1/9.1/9.4
Leaves area (10 plants per tray)	cm <sup>2</sup>	0/0/0	0.49/0.50/0.51	0.62/0.61/0.62
<b>Determining fresh biomass yield and dry matter content</b>				
Fresh weight	g tray (661 cm <sup>2</sup> )	0/0/0	48.45/47.84/48.32	58.45/59.12/58.89
Dry weight	g tray (661 cm <sup>2</sup> )	0/0/0	3.35/3.31/3.35	4.05/4.10/4.08
Total phenols	mg kg <sup>-1</sup>	0/0/0	99/98/97	114/101/107

Note: \*r1- top tray; r2 - middle tray; r3 - bottom tray

### Fresh weight accumulation in semi-controlled settings

The amount of fresh weight (FW) depends on both the cultivated species and the variety (Table 4), respectively its suitability for hydroponic microgreens. Among the two tested species, lettuce provides a higher average FW production, respectively 809.56 g m<sup>-2</sup>. In the case of basil, the best production of FW microgreens was ensured by Germaline Basilic Bio, 783.54 g m<sup>-2</sup>, being classified by the Duncan test (b). The best amount of FW is provided by lettuce - *Lolla Rossa*, with 889.86 g m<sup>-2</sup>.

**Table 4.** The amount of fresh weight accumulation in the two species (basil and lettuce) and varieties in semi-controlled conditions

Species	Variety	Fresh weight		Difference ±	Significance
		g m <sup>-2</sup>	%		
<b>Basil</b>	Control	734.73	100	Ct.	Ct.
	<i>Germaline Basilic Bio</i>	783.54 b	106.60	48.81	*
	<i>Grand Vert</i>	685.92 a	93.40	-48.81	0
<b>Lettuce</b>	Control	809.56	100	Ct.	Ct.
	<i>Little Gem</i>	729.25 a	90.10	-80.31	00
	<i>Lolla Rossa</i>	889.86 b	109.90	80.31	**

Note: Basil: DL (p 5%)= 28.03 g m<sup>-2</sup>; DL (p 1%)= 64.73 g m<sup>-2</sup>; DL(p 0.1%)= 205.98 g m<sup>-2</sup>; Ct.- control. Lettuce: DL (p 5%)= 24.08 g m<sup>-2</sup>; DL (p 1%)= 55.61 g m<sup>-2</sup>; DL(p 0.1%)= 176.96 g m<sup>-2</sup>.

### Dry weight accumulation in semi-controlled settings

Following the statistical processing of the data regarding dry weight (DW) accumulation, it is found that the results differ from fresh weight (Table 5). The results obtained on the repetitions (GoHydro platform trays) were processed statistically and interpreted in relation to the average of the species. Basil provides the highest amount of DW, the average of the species being 73.36 g m<sup>-2</sup>. The same results after processing the data with the Duncan test, respectively the two varieties of basil ensure the highest DW results.

**Table 5.** The amount of dry weight accumulation in the two species (basil and lettuce) and varieties in semi-controlled conditions

Species	Variety	Dry weight		Difference ±	Significance
		g m <sup>-2</sup>	%		
<b>Basil</b>	Control	73.36	100	Ct.	Ct.
	<i>Germaline Basilic Bio</i>	78.26 b	106.70	4.90	**
	<i>Grand Vert</i>	68.45 a	93.30	-4.90	00
<b>Lettuce</b>	Control	56.08	100	Ct.	Ct.
	<i>Little Gem</i>	50.48 a	50.00	-5.60	00
	<i>Lolla Rossa</i>	61.67 b	110.00	5.60	**

Note: Basil: DL (p 5%)= 2.04 g m<sup>-2</sup>; DL (p 1%)= 4.70 g m<sup>-2</sup>; DL(p 0.1%)= 14.96 g m<sup>-2</sup>; Ct.- control. Lettuce: DL (p 5%)= 1.72 g m<sup>-2</sup>; DL (p 1%)= 3.97 g m<sup>-2</sup>; DL(p 0.1%)= 12.64 g m<sup>-2</sup>.

### Total Phenolics/Morphology in semi-controlled settings

The results obtained in the case of total phenols are presented in the Table 6. The variability of the data recorded for total phenols is very high, the statistical processing does not highlight results with a significant difference. Lettuce contains 98-107.33 mg kg<sup>-1</sup>, and basil 141-186.33 mg kg<sup>-1</sup> total phenols.

From the morphological analysis of plant development, both in basil and in lettuce, no disease attack is registered, due to the maintenance of the health of the water in the feed basin and a quantity of around 6.4-6.5 mg L<sup>-1</sup> of dissolved oxygen. An exception is made in the case of lettuce, the Paris White variety, which registers until the end,

grade 3 in the degree of attack. This is due exclusively to this variety in which the seed had an inadequate quality for the production of hydroponic microgreens. There are differences regarding the number of leaves, the length of the microgreens, etc. but these are given in particular by the variety used. Within the same variety, the differences between repetitions were very small, insignificant.

**Table 6.** The amount of total phenols in the two species (basil and lettuce) and varieties in semi-controlled conditions

Species	Variety	Total phenols		Difference ±	Significance
		mg kg <sup>-1</sup>	%		
<b>Basil</b>	Control	163.67	100	Ct.	Ct.
	<i>Germaline Basilic Bio</i>	141.00 a	86.20	-22.67	-
	<i>Grand Vert</i>	186.33 a	113.80	22.67	-
<b>Lettuce</b>	Control	102.67	100	Ct.	Ct.
	<i>Little Gem</i>	98.00 a	95.50	-4.67	-
	<i>Lolla Rossa</i>	107.33 a	104.50	4.67	-

Note: Basil: DL (p 5%)= 235.94 mg kg<sup>-1</sup>; DL (p 1%)= 544.86 mg kg<sup>-1</sup>; DL(p 0.1%)= 1733.91 mg kg<sup>-1</sup>; Ct.- control. Lettuce: DL (p 5%)= 14.96 mg kg<sup>-1</sup>; DL (p 1%)= 34.56 mg kg<sup>-1</sup>; DL(p 0.1%)= 109.97 mg kg<sup>-1</sup>.

### Parameters collected related to the growth environment in operational settings

The growing conditions of the two species of microgreens (basil and lettuce) it is specific to the conditions in the operational environment, being also influenced by the external environmental conditions, respectively especially by cloudiness. Thus, a temperature was kept in the atmosphere with a oscillation of 3-7 °C, an atmospheric humidity oscillation of 8-15 %, and respectively a variable amount of light (of 135-1645 μmol m<sup>-2</sup> s<sup>-1</sup>).

The irrigation water had a constant temperature of 19-21 °C, a pH of 6.8-7, an electrical conductivity between 1.4-1.5 dS m<sup>-1</sup>, and respectively a quantity of 6.4-6.5 mg L<sup>-1</sup> dissolved oxygen. The images of the two experiments are presented in Figure 3. Parameters related to microgreen growth in operational settings, production and quality in the basil experiment is presented in Table 7, and for lettuce in Table 8.



**Figure 3.** Experiment with seed of the basil (a), basil after 7 days from sowing (b), lettuce after 14 days from sowing (c) in operational environmental conditions

**Table 7.** Parameters related to microgreen growth, production and quality in the basil experiment in operational conditions

Parameter	Unit of measurement	Day 18/21.11.2022	
		<i>Germaline</i>	<i>Grand Vert</i>
<b>Determining the health state of plants</b>			
Intensity of the disease attack	Note 1-9	1	1
<b>Measurements on the morphology of plants*</b>		r1/r2/r3	r1/r2/r3
Stem length	mm	35.2/34.1/31.1	28.4/28.2/25.2
Internode length	mm	35.2/34.1/31.1	28.4/28.2/25.2
Number of leaves	no	2/2/2	2/2/2
Number of plants	no cm <sup>-2</sup>	10.3/9.8 /10.1	9.6/8.9/8.6
Leaves area (10 plants per tray)	cm <sup>2</sup>	0.61/0.59/0.58	0.52/0.53/0.50
<b>Determining fresh biomass yield and dry matter content</b>			
Fresh Weight	g tray (992 cm <sup>2</sup> )	89.41/87.75/85.74	77.41/75.42/74.56
Dry Weight	g tray (992 cm <sup>2</sup> )	9.01/8.85/8.74	7.84/7.67/7.56
Total phenols	mg kg <sup>-1</sup>	106/109/108	109/127/118

Note: r1- top tray; r2 - middle tray; r3 - bottom tray

**Table 8.** Parameters related to microgreen growth, production and quality in the lettuce experiment in operational conditions

Parameter	Unit of measurement	Day 15	Day 15/19.12.2022	
		/19.12.2022	<i>Paris White</i>	<i>Little Gem</i>
<b>Determining the health state of plants</b>				
Intensity of the disease attack	Note 1-9	4	1	1
<b>Measurements on the morphology of plants*</b>		r1/r2/r3	r1/r2/r3	r1/r2/r3
Stem length	mm	0/0/0	31.4/31.8/32.4	32.4/31.9/32.8
Internode length	mm	0/0/0	31.4/31.8/32.4	32.4/31.9/32.8
Number of leaves	no	0/0/0	2.5/2.4/2.6	2.3/2.1/2.2
Number of plants	no cm <sup>-2</sup>	0/0/0	10.4/10.2/10.3	9.6/9.5/9.8
Leaves area (10 plants per tray)	cm <sup>2</sup>	0/0/0	0.51/0.49/0.50	0.61/0.59/0.64
<b>Determining fresh biomass yield and dry matter content</b>				
Fresh Weight	g tray (661 cm <sup>2</sup> )	0/0/0	47.24/48.21/47.54	54.12/55.84/55.45
Dry Weight	g tray (661 cm <sup>2</sup> )	0/0/0	3.27/3.34/3.29	3.75/3.87/3.84
Total phenols	mg kg <sup>-1</sup>	0/0/0	98/101/97	104/101/105

Note: r1- top tray; r2 - middle tray; r3 - bottom tray



### Fresh weight accumulation in operational settings

The amount of fresh weight (FW) depends on both the cultivated species and the variety, respectively its suitability for hydroponic microgreens (Table 9). Among the two species tested, basil ensures a better average FW production, respectively 823.74 g m<sup>-2</sup>. The best amount of FW is provided by Basil - *Germaline Basilic Bio*, with 883.40 g m<sup>-2</sup>.

**Table 9.** The amount of fresh weight accumulation in the two species (basil and lettuce) and varieties in operational conditions

Species	Variety	Fresh weight		Difference ±	Significance
		g m <sup>-2</sup>	%		
Basil	Control	823.74	100	Ct.	Ct.
	<i>Germaline Basilic Bio</i>	883.40 b	107.20	59.66	**
	<i>Grand Vert</i>	764.08 a	92.80	-59.66	00
Lettuce	Control	777.61	100	Ct.	Ct.
	<i>Little Gem</i>	721.08 a	92.70	-56.53	00
	<i>Lolla Rossa</i>	834.14 b	107.30	56.53	**

Note: Basil: DL (p 5%)= 14.81 g m<sup>-2</sup>; DL (p 1%)= 34.21 g m<sup>-2</sup>; DL(p 0.1%)= 108.87 g m<sup>-2</sup>; Ct.- control. Lettuce: DL (p 5%)= 20.00 g m<sup>-2</sup>; DL (p 1%)= 46.18 g m<sup>-2</sup>; DL(p 0.1%)= 146.96 g m<sup>-2</sup>.

### Dry weight accumulation in operational settings

Following the statistical processing of the data regarding dry weight (DW) accumulation, it is found that the basil (Table 10) provides the highest amount of DW (83.45 g m<sup>-2</sup>). The best production DW microgreens was obtained at *Germaline Basilic Bio*, respectively 89.38 g m<sup>-2</sup>, with a distinctly significantly positive difference compared to the control (average).

**Table 10.** The amount of dry weight accumulation in the two species (basil and lettuce) and varieties in operational conditions

Species	Variety	Dry weight		Difference ±	Significance
		g m <sup>-2</sup>	%		
Basil	Control	83.45	100	Ct.	Ct.
	<i>Germaline Basilic Bio</i>	89.38 b	107.10	5.93	***
	<i>Grand Vert</i>	77.52 a	92.90	-5.93	000
Lettuce	Control	53.86	100	Ct.	Ct.
	<i>Little Gem</i>	49.92 a	92.70	-3.93	00
	<i>Lolla Rossa</i>	57.79 b	107.30	3.93	**

Note: Basil: DL (p 5%)= 0.13 g m<sup>-2</sup>; DL (p 1%)= 0.30 g m<sup>-2</sup>; DL(p 0.1%)= 0.95 g m<sup>-2</sup>; Ct.- control. Lettuce: DL (p 5%)= 1.36 g m<sup>-2</sup>; DL (p 1%)= 3.13 g m<sup>-2</sup>; DL(p 0.1%)= 9.97 g m<sup>-2</sup>.

### Total Phenolics/Morphology in operational settings

The results obtained in the case of total phenols are presented in the Table 11. The variability of the data recorded for total phenols is very high, the statistical processing does not highlight results with a significant difference. Lettuce contains 98.67-103.33 mg kg<sup>-1</sup>, and basil 107.67-118 mg kg<sup>-1</sup> total phenols.

From the morphological analysis of plant development, both in basil and in lettuce, no disease attack is registered, due to the maintenance of the health of the water in the feed basin and a quantity of around 6.4-6.5 mg L<sup>-1</sup> of dissolved oxygen. An exception is made in the case of lettuce, the Paris White variety, which registers until the end, grade 4 in the degree of attack. Due to the higher thermal oscillations than in the case of the experiment under controlled conditions, the degree of attack was slightly higher. This is due exclusively to this variety in which the seed had an inadequate quality for the production of hydroponic microgreens. There are differences regarding the

number of leaves, the length of the microgreens, etc. but these are given in particular by the variety used. Similar to the experiment in semi-controlled conditions and in the case of the experiment in operational conditions within the same variety, the differences between repetitions were very small, insignificant.

**Table 11.** The amount of total phenols in the two species (basil and lettuce) and varieties in operational conditions

Species	Variety	Total phenols		Difference ±	Significance
		mg kg <sup>-1</sup>	%		
Basil	Control	112.03	100	Ct.	Ct.
	<i>Germaline Basilic Bio</i>	107.67 a	95.40	-5.17	-
	<i>Grand Vert</i>	118.00 a	104.60	5.17	-
Lettuce	Control	101.00	100	Ct.	Ct.
	<i>Little Gem</i>	98.67 a	97.70	-2.33	-
	<i>Lolla Rossa</i>	103.33 a	102.30	2.33	-

Note: Basil: DL (p 5%)= 18.63 mg kg<sup>-1</sup>; DL (p 1%)= 43.03 mg kg<sup>-1</sup>; DL(p 0.1%)= 136.93 mg kg<sup>-1</sup>; Ct.- control. Lettuce: DL (p 5%)= 10.34 mg kg<sup>-1</sup>; DL (p 1%)= 23.87 mg kg<sup>-1</sup>; DL(p 0.1%)= 75.96 mg kg<sup>-1</sup>.

## DISCUSSION

Feedback from these tests will be used for final validation of the hydroponic platform components. Thus, it is very important to analyze the factors that influenced the production results and the quality of microgreens (Parkes et al., 2022).

First of all, it should be mentioned that all cultures had a very variable density, due to the unknown percentage of germination. A standard germination of 90% was considered. The amount of seed on the tray was determined with the Francesco Di Gioia Microgreens Seed Calculator. However, we found in the end that actual germination was about 60-70%. As a result, a first recommendation is the need to know/determine the actual germination of the seeds, before cultivating microgreens as Galieni et al., 2020, also recommends. However, it should be mentioned that many seeds, including those intended for the production of microgreens, did not mention the germination capacity (Palmitessa et al., 2022).

Then, it should be mentioned that seeds were moistened differently. Thus, we consider that it is necessary to wet the seeds in advance, for a uniform development of microgreens. Nevertheless, Li et al, 2021, consider that use of pre-sowing seed soaking treatment to advance seed germination should be weighed against its possible effects in reducing microgreen yield and mineral nutrient concentrations.

Microgreens attacked by pathogens were identified. Pathogenic agents can result from the seed used or the substrate used and can spread easily due to the proximity of the seedlings (Dhawi, 2023). A solution can be the one recommended by Tavan et al., 2021, namely that seeds must first be sterilized by soaking for two minutes in 80% ethanol, rinsed twice with distilled water and then dried in an oven at 45°C for 40 minutes. The use of hemp substrate in the hydroponic production of microgreens can be used successfully, as shown by Li et al., 2021, but we believe that special attention must be paid to sanitary conditions.

In semi-controlled conditions, the average fresh weight production achieved by the two species (and four varieties) is 734.73 g m<sup>-2</sup> for basil, 809.56 g m<sup>-2</sup> for lettuce, and represents 823.74 g m<sup>-2</sup> for basil, 777.61 g m<sup>-2</sup> for lettuce in operational environmental conditions. The results can be explained by the light oscillation between 140-1645 μmol m<sup>-2</sup> s<sup>-1</sup> in operational conditions and a constant intensity of 1217 μmol m<sup>-2</sup> s<sup>-1</sup> in semi-controlled conditions with a photoperiod of 12/12 h. The natural intensity of the light, even oscillating, has a good effect on the development of microgreens. At the same time, it seems that low light intensity (alternation with high intensity) promotion on elongation growth might be a phenomenon in operational environmental conditions, which could be beneficial to microgreens (Gao et al., 2021). Thus, we can find that the internode length in operational environmental conditions varies for basil between 25.2-35.2 mm, for lettuce between 31.9-32.8 mm, and in semi-controlled environmental conditions, it varies for basil between 25.2-32.1 mm and for lettuce between 30.2-31.9 mm.

Microgreens can be successfully grown in operational (Langenfeld et al., 2022) and semi-controlled environmental conditions (Parkes et al., 2022), identified by controlling specific environmental parameters (Avgoustaki et al., 2020) such as air temperature, relative humidity, substrate, water and light incidence and others. These comparative tests can be used to carry out detailed analyzes by varying some experimental factors, such as the seeding density, the management of environmental conditions (Samuoliene et al., 2012; Sabzalian et al., 2014; Brazaityte et

al., 2016) and, respectively, the improvement of IT management programs (Lobiuc et al., 2017) or platforms used for the production of microgreens in one's own kitchen.

## CONCLUSIONS

Despite the short growth cycle, in the production of microgreens, special attention must be paid to the control of the growth environments, which is one of the most important factors in the production process, influencing the quantity and quality of microgreens.

**In semi-controlled settings:** the growing conditions of the two species of microgreens (basil and lettuce) are specific to semi-controlled conditions, using the Eden Grow plant growing tent. Thus, a temperature was kept in the atmosphere with a maximum oscillation of 3-4 °C, an atmospheric humidity oscillation of 8-10%, and respectively a constant amount of light (of 1217  $\mu\text{mol m}^{-2}\text{s}^{-1}$ ). The irrigation water had a constant temperature of 19-21 °C, a pH of 6.8-7, an electrical conductivity between 1.4-1.5  $\text{dS m}^{-1}$ , and respectively a quantity of 6.4-6.5  $\text{mg L}^{-1}$  dissolved oxygen. Among the two tested species, lettuce provides a higher average FW production, respectively 809.56  $\text{g m}^{-2}$ . In the case of basil, the best production of FW microgreens was ensured by *Germaline Basilic Bio*, 783.54  $\text{g m}^{-2}$ . The best amount of FW is provided by lettuce - *Lolla Rossa*, with 889.86  $\text{g m}^{-2}$ . The variability of the data recorded for total phenols is very high, the statistical processing does not highlight results with a significant difference. Lettuce contains 98-107.33  $\text{mg kg}^{-1}$ , and basil 141-186.33  $\text{mg kg}^{-1}$  total phenols.

**In operational environments settings:** the growing conditions of the two species of microgreens (basil and lettuce) are specific to the conditions in the operational environment, being also influenced by the external environmental conditions, for example especially by cloudiness. Having supplementary light, depending on time of year and degree of cloudiness is therefore an important consideration. Among the two species tested, basil ensures a better average FW production, respectively 823.74  $\text{g m}^{-2}$ . The best amount of FW is provided by Basil - *Germaline Basilic Bio*, with 883.40  $\text{g m}^{-2}$ . Basil provides the highest amount of DW (83.45  $\text{g m}^{-2}$ ). The best production DW microgreens was obtained at *Germaline Basilic Bio*, respectively 89.38  $\text{g m}^{-2}$ , with a distinctly significantly positive difference compared to the control (average).

**From the morphological analysis of plant development,** both in basil and in lettuce, no disease attack is registered, due to the maintenance of the health of the water in the feed basin and a quantity of around 6.4-6.5  $\text{mg L}^{-1}$  of dissolved oxygen. An exception is made in the case of lettuce, the *Paris White* variety, which registered a grade 3 (semi-controlled settings) and 4 (operational environments settings) in the degree of attack until harvest. Due to the higher thermal oscillations than in the case of the experiment under controlled conditions, the degree of attack was slightly higher. This is due exclusively to this variety in which the seed had an inadequate quality for the production of hydroponic microgreens. The conclusions of the experiment, which require the optimization of production procedures with the GoHydro platform, are related to the type of substrate used, the density of the microgreens culture, the attack by pathogens and the seed germination procedure.

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