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PhD thesis

EXPERIMENTAL STUDY OF A MINI AND STANDARD SOILLESS CULTIVATION SYSTEM IN CONTROLLED ENVIRONMENT AGRICULTURE

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ABSTRACT

Agricultural innovations and information technologies have introduced promising cultivation techniques leading to an evolution of the simple greenhouse to a high-tech Controlled Environment Agriculture (CEA). These facilities fall within the concept of Urban Agriculture. Enclosing the crop within an envelope has a beneficial effect on the microclimate, and against unfavourable outdoor climate conditions and pests. However, there are still challenges to overcome for an efficient and viable system. Improvement of the cultivation system and of its operation to enhance resources usage and use of natural energy is essential. The development of a mini hydroponic cultivation system and its characterization in a controlled environment fits well with this purpose.

In the first section, the operation system of hydroponic cultivation is described, starting from the general aspect of this topic and, then, focusing on the technical aspects (materials and main environmental factors). This was done not only for well comprehension of the context and of successive explanation of the thesis experimental field, but also because it reason for deepening to carry the work out. The development of a controlled mini-environment equipped with a specially designed hydroponic above-ground cultivation system represented the major challenge that requires the design of both the hydroponic system and the controlled environment. In addition, the solution must include a set of probes and a supervisory system to properly control the main parameters of interest.

After the conceptualization of the experimental apparatus, in the second section it is reported a further experimental work conducted in a large standard hydroponic greenhouse for the evaluation and characterization of microclimatic parameters. Test were conducted in two different growing cycle of lettuce crop. This work represented an useful point for the project due to the fact that permitted to analyse personally hydroponic systems from a large scale point of view.

In the third section, the focus moves from the large-scale to the small-scale hydroponics. To this aim, the experimental environment chosen for the tests was a laboratory of 40 m^3, that represented a functional example of Controlled Cultivation Environment. At first, it was evaluated the distribution of microclimatic parameters in the confined and conditioned environment. Tests point-by-point were conducted dividing the cultivation

room uniformly and setting a microclimatic control unit with parameters for simulating a lettuce seedling.

After the characterization and the collection of physical quantities, in the fourth section is described the development of an hydroponic drip system in less 1 m^3 with artificial light and in a fully closed environment. This system, placed in the previous tested confined environment, represents an interesting solution of mini hydroponic. Microclimatic monitoring of the system and also of agronomic parameters of lettuce plants cultivated was done. The final data were elaborated considering also data trends get from the large-scale greenhouse.

A further paragraph concerns additional technologies developed that can find applications in this project, as a result of the doctoral experience abroad.

Finally, section 5 reported the main conclusions of the present work.

1 Introduction

According to projections from the United Nations Food and Agriculture Organization (FAO) the human population is expected to reach about 9 billion people by the year 2050 a significant tipping point in history heritage [1]. Food production will need to increase globally up about 70% from 2007 levels in order to meet the demand. In addition due to global urbanization it is estimated that 75% of the world's population would reside in cities by 2050. [2].

Today the shortage and/or unstable supply of food, the shortage of resources, and the lack of arable land [3] increasingly require the introduction of new technologies in agriculture in order to reduce energy consumption and environmental degradation compared to current plant production systems. Indeed, due to rapid urbanization and industrialization not only the cultivable land is decreasing, but also conventional agricultural practices are causing a wide range of negative impacts on the environment. In this sense agricultural innovations and information technologies (IT) have introduced promising cultivation techniques leading to an evolution of the simple greenhouse to a high-tech Controlled Environment Agriculture (CEA) [4].

CEA refers to innovative methods for controlling plant growth and development by exploiting advanced techniques and innovations in technology [3] and it pertains to the concept of Urban Farming o Urban Agriculture (UA).

Enclosing the crop within an envelope with artificial lights leads to benefits that include: positive effects on the microclimate, high resource use efficiency, high annual productivity per unit of land area, and production of high-quality plants without using pesticides [5, 6].

In this context, hydroponic systems represent an interesting solution. Indeed, the most promising technology of the soilless cultivation inside greenhouse and hydroponics systems, in which soil is replaced by water-based mineral nutrient solutions in aqueous solvents, are now very common and efficient [7]. Hydroponics represents a sustainable alternative for urban and peri-urban areas and contribute to the sustainable development goal (SDG) number 11 (Sustainable Cities and Communities) of the United Nations 2030 Agenda for Sustainable Development [8].

The increasing use of hydroponics is important for Urban Agriculture and a breakthrough in the food industry's supply chain society's well-being and the environment. However, in order to achieve its maximum potential it must enter the world of small- and medium-

scale farming [8]. To maximize cost-benefit at small- and medium-scale levels producers need understanding the advantages of using new technologies. This is done by having a specific knowledge of the fundamentals of hydroponics, so that appropriate technologies can be developed. In particular, attention needs to be given to the key technical aspects in order to develop an advanced system that operates successfully. This type of research includes technical solutions and scientific analyses for the production of plants, handling of environmental parameters machines, for material management, process control, monitoring, and collection of information to implement decisional systems [9].

On the other hand, it is also important to consider crops that are suitable for this kind of system. Leafy vegetables are increasingly becoming common in hydroponics, especially lettuce that is the largest hydroponically grown vegetable in indoor farming. Indeed, it is predicted that throughout the forecast period the worldwide lettuce market would grow at the quickest CAGR (Compounded Average Growth Rate) of more than 23% [10].

1.1 Research Project

The present Ph.D. thesis deals with the research project of a fully closed soilless cultivation system for lettuce with artificial light and with particular attention to its management in a small-scale.

Main purpose of this research project was the development of a reliable control logic of the system acting on parameters like temperature relative humidity air flow light nutrients CO_2 water and other relevant agronomic variables. Thanks to a network of sensors which can work either offline or cloud-based way the system was continuously (real-time) monitored.

Other essential aspect was the choice of the cultivation technique and the identification of the target: hydroponic system in small-scale environment. This aim was achieved contextualizing the hydroponic system with a preliminary study of the main microclimatic parameters in a basic large hydroponic greenhouse comparing the case study without a microclimatic controlling system and with a ventilation system. The collected data were useful as a comparison with the monitoring tests done in the controlled environment agriculture object of the thesis.

Then the discussion moved towards the small-scale implant. Initially, it was necessary to characterize the chosen experimental environment with physical quantities point-by-point thanks to a set of probes. After determining the goodness of the environment, it was

conceptualized and realized in it a mini hydroponic system and conducted the cycle cultivation system of lettuce.

1.2 Hydroponics

The practice of growing vegetables without the use of soil [11]. In hydroponics plants are grown without soil by giving them nutrient-rich water solvent solutions this latter normally gained from the soil in traditional farming. The basic goal of hydroponics is to provide the best nutritional environment for the best plant growth and yield which is further enhanced by controlling the climate.

1.2.1 Background

The term Hydroponics was derived from the Greek words ὑδρο- (hýdro-) from ὕδωρ (húdōr "water") and πόνος (pónos "work, labour" and literally means water work. Hydroponics is a type of horticulture a method that uses nutrient mineral solutions instead of tillage [12]. The murals on the walls of the more than 4000-year-old Egyptian temple Deir El Bahari are among the earliest evidences of hydroponics. [13].

Hydroponics was adopted to grow mainly ornamental plants in Babylon during the sixth century BCE [14]. The Mexican Aztec culture created the chinampa in pre-Columbian America between the X and XI centuries CE to cultivate crops on the lake beds flats in the Valley of Mexico and it is thought that Mesoamericans used it as well. [15]. In 1600 Jean Baptiste Van Helmont a Belgian scientist conducted a series of experiments to demonstrate that plants can get some nutrients from water.

German botanists made great progress in soilless growing techniques between 1859 and 1865. William Frederick Gerick promoted solution culture for agricultural crop production in 1929. However the word "Hydroponic" was firstly introduced by the William Frederick Gerick in 1937. Commercial hydroponics farms were developed all over the world between 1960 and 1970. The 1980s saw the development of numerous high-tech automated hydroponic farms all over the world.

1.2.2 General aspect

Hydroponics is a cultivation system that does not require soil to grow plants. In this technique plants are grown either on natural or artificial substrates where the roots easily extract the nutrients from a prepared nutrient solution. In general, there are two main hydroponics techniques: media culture method and solution culture method. Table 1

below compares the effectiveness of both methods in terms of percentage irrigation water savings percentage fertilizer efficiency percentage productivity growth and percentage water productivity [16-21].

***Tab. 1**: Comparison between hydroponic technique.*

Parameters	Hydroponic Technique	
	Solution Culture	
	Open	Close
% of irrigation water saving	85	90
% of fertilizer saving	68	85
% of productivity increase	200	300
% of water productivity	2000	3500

In particular, in solution culture method plants are grown in solution culture while directly suspending their roots in nutrient solution [22]. It can also be divided into the following systems: Deep Water Culture, Drip System, Nutrient Film Technique (NFT), Ebb and flow, Aeroponics, and Aquaponics (for details see Figure 1). This differentiation in each method is based on the structure set up.

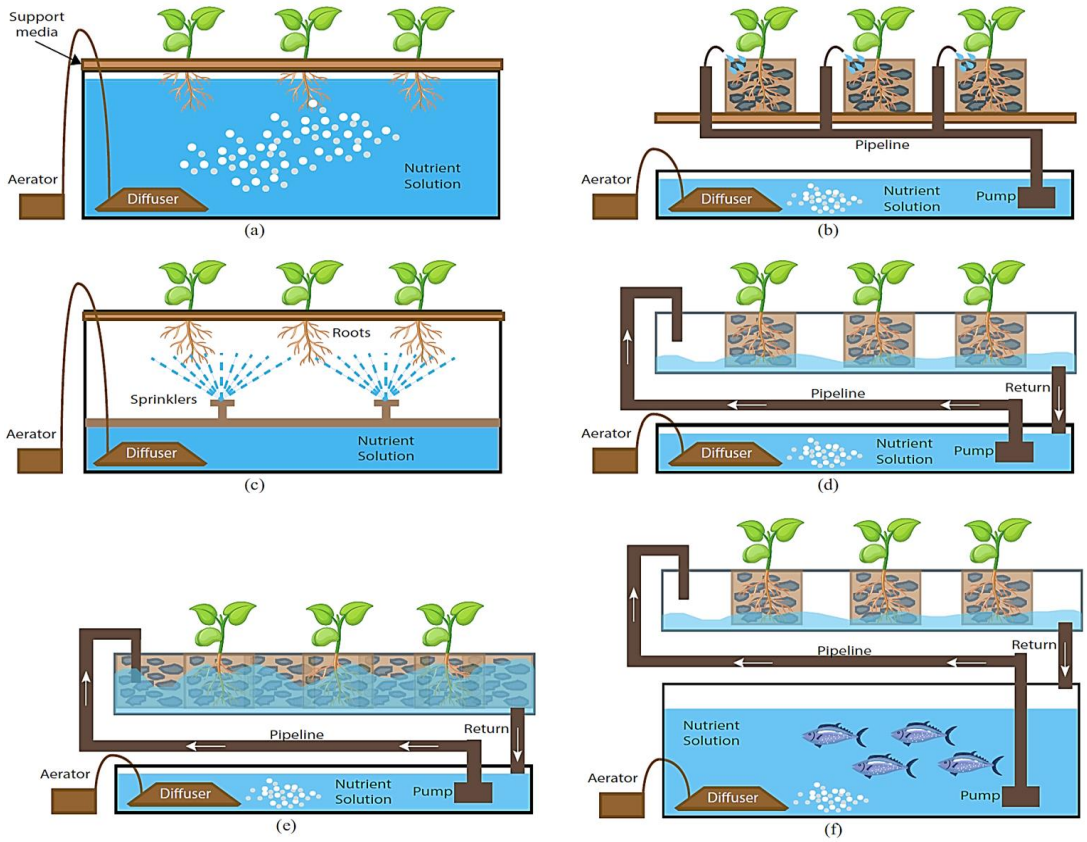


Fig. 1 *Different types of hydroponic systems: a) Deep Water Culture; b) Drip System; c) Aeroponics; d) Nutrient Film Technique (NFT); e) Ebb and flow; f) Aquaponics.* *(From: Velazquez-Gonzalez R. S. Garcia-Garcia A. L. Ventura-Zapata E. Barceinas-Sanchez J. D. O. & Sosa-Savedra J. C. (2022). A review on hydroponics and the technologies associated for medium-and small-scale operations. Agriculture 12(5) 646).*

1.3 Cultivation Technique

1.3.1 Floating Root System or Deep Water Culture (DWC)

In this technique (Figure 1a) plants are placed in net pots above water level but the roots are immersed in the nutrient solution. In this system it is fundamental to monitor the oxygen and nutrient concentrations pH and salinity [23]. Due to the system's restricted air-water exchange area in relation to the volume of the solution and the low oxygen diffusion coefficient in the water hypoxic conditions at the root level is frequently developed. Recirculating deep water culture systems (RDWC) which employ a reservoir to supply nutrient solution to numerous buckets are used to get over this restriction by oxygenating the nutrient solution with air pumps or by using other methods.

1.3.2 Drip Irrigation System

Here drip tubes convey a precise quantity of nutrient solution that thanks to a pump is provided to the roots [24] (Figure 1b), greatly increasing the water use efficiency of the crop.

At regular intervals the solution is applied and the remaining solution is added back to the storage tank. The used nutrient solution in continuous drip systems can either be returned to the tank or run off as waste depending on whether they are recovery or non-recovery systems. Recovery systems are more economical because they utilize the nutrient solution more efficiently while nonrecovery systems need less maintenance because the pH balance and nutritional strength are constant with the supply of fresh solution.

1.3.3 Aeroponics

The plants grow with their root systems suspended in an intermittent or continuous ultrafine mist of nutritional solution. The solution is sprayed frequently by a system of nozzles (Figure 1c). The main advantage of this technique is that it does not need an airing system since oxygen is carried in the nutrient solution that is sprayed. With the support of polyurethane foam plants are held in place in holes in polystyrene panels that are placed over a metal frame in a horizontal or inclined configuration. Closed containers with a square or triangular section are set on the panels.

1.3.4 Nutrient Film Technique (NFT)

This technique (Figure 1d) provides for the cultivation of plants in channels without any substrate and a slightly sloping (1.5-2.5%) within which flows a thin film of nutrient solution with a flow of 1-3 L/min. It is a closed loop system and the nutrient solution is pumped into the upper part of the channel and flows by gravity to the collection pipes. The flow of nutrient solution can be continuous or discontinuous and the excess solution returns to the tank by gravity. The thin water stream (1-2 mm deep) ensure that the roots receive enough oxygen because the dense root mat that forms on the channel’s bottom is constantly exposed to the air on its upper surface.

Although NFT needs less nutrient solution than the floating root system still it requires more energy and materials to function.

1.3.5 Ebb and Flow

In this system plants are placed in a tray filled with substrate (perlite, rockwool, or enlarged clay pebbles) consists of a tray filled with substrate that is periodically submerged by the nutrient solution pumped from a tank below. By gravity the water returns to the tank and it is reused (Figure 1e).

1.3.6 Aquaponics

Aquaponics is a type of plant growing system that combines aquaculture and hydroponic cultivation in order to obtain a symbiotic environment. The recycling of water from the fish tank is made possible by the plants uptake of nutrients and the microbial nitrification and denitrification processes. This creates a well-functioning microecosystem (Figure 1f).

1.4 Advantages and Disadvantages

Hydroponics, as other agriculture technique, presents advantages and disadvantages. The opportunity to practice hydroponics in difficult areas and anywhere that a controlled environment can be established is one of the advantages. Furthermore, depending on the type of plant, it is possible to devise vertical arrangements for increased produce output. Compared to crops grown using traditional systems crops without soil normally allow greater certainty increasing the yield per cycle per crop. The factors that positively influence these two aspects are: the absence of pathogens in the growing medium; the absence of competition between plants at the level of nutrition; the best conditions of the controlled environment and therefore less or total absence frequency of phytopathogenic attacks. Additionally, as nutrition is given in accordance with the plant's physiological needs the quality of the products is guaranteed. However, it is important to keep in mind that in completely automated greenhouses even little modifications to the operational environment could cause immediate crop reactions.

Hydroponic practice demonstrates positive factors also from a sustainability point of view: in particular, this is made possible by the possibility of the nutrient solution to be recycled and the absence of soil. In this way water evaporation, leaching, seepage, and pollution are minimized. However if the residual nutrient solution is not properly disposed of the discharged solution rich in phosphate and nitrates can cause an excessive development of algae and other microorganisms in river systems and effluents leading to major environmental issues.

Among drawbacks hydroponics certainly has high initial cost due to the cost of required equipment for the operations. Moreover hydroponics could require for more technical expertise generally involving a set of sensors and tools for an accurate follow-up of the crop status.

1.5 Hydroponic cultivation components

1.5.1 Growing media

A substrate is the physical medium that supports plants by the stem and keep them under appropriate growing conditions [25]. The most common growth media are substrates inorganic (sand, gravel, perlite, rockwool, volcanic stones etc.) or organic (peat, bark, coir, rice hulls etc.). However, other types of substrates are used when the choice is strongly influenced by availability and economic cost. During the choice of the medium there are some relevant factors to take in account [26-28]: adequate mechanical properties to ensure the stability of the plant; high porosity (at least 75%); an adequate distribution of air i.e. oxygen and water an adequate balance between macro and microporosity to ensure a good water capacity and at the same time facilitate gas exchanges in the hypogeal part of the plant; low soluble salt content; low cation and anion exchange capacity; ability to maintain original characteristics for crops with a long growing cycle; chemically inert; biologically inert.

In particular, successful hydroponic cultivation depends on root aeration. The management of substrate-grown crops must take into account the variables affecting air availability in growing media. In media with low air-filled porosity oxygen deficit is easily possible especially if the plants have rapid growth and intense root respiration. The optimal porosity is of 75 % and with the right balance between micro and macro pores of 40- 60 % and 15-35 %, respectively. For production in small containers the total pore space is expected to reach 85 % of the volume [29].

1.5.2 Crops in Hydroponics

Protected cultivation with hydroponic greenhouse cultivation is the only method for creating a condition that is suitable for greater crop yield. Thanks to hydroponics it is possible to cultivate a considerable variety of crops: vegetables, flowers, cereals, fruits, fodder, condiments, and medicinal plant [30]. Table 2 shows the summary of these crops grown in hydroponics. According to Singh and Singh, 2012 [30] hydroponic greenhouse

yield per area is greater than open agriculture yield per area. Table 3 shows the yield comparison between hydroponic and open agriculture. This comparison showed that there is big difference between this two methods due to controlled environment in case of hydroponic greenhouse cultivation and the re-use of nutrient solution.

***Tab. 2**: Summary of Crops for Hydroponic Greenhouse Cultivation [30].*

Type of Crops	Name of Crops	
	Common Name	Botanical Name
Cereals	Rice	Oryza sativa
	Maize	Zea mays
	Wheat	Triticum aestivum
	Oat	Avena sativa
	Soybean	Glycin max
	Peas	Pisum sativum
Vegetables	Tomato	Lycopersicon lycopersicum
	Chilli	Capsicum frutescens
	Brinjal	Solanum melongena
	Green bean	Phaseolus vulgaris
	Bell pepper	Capsicum annum
	Beet	Beta vulgaris crassa
	Potato	Solanum tuberosum
	Cabbage	Brassica oleracea var.
	Cauliflower	Brassica oleracea
	Cucumber	Cucumis sativus
	Onion	Allium cepa
	Radish	Raphanus sativus
	Lettuce	Latuca sativa
Fruits	Strawberry	Fragaria ananassa
	Melons	Cucumis melo
Fodder crops	Sorghum	Sorghum bicolor
	Alfalfa	Cynodon dactylon
	Barley	Hordeum vulgare
	Bermuda grass	Cynodon dactylon
	Carpet grass	Axonopus compressus
Flower	Marigold	Tagetes patula
	Roses	Rosa berberifolia
	Carnations	Dianthus caryophyllus
	Chrysanthemum	Chrysanthemum indicum
Condiments	Parsley	Petroselinum crispum
	Mints	Mentha spicata
	Sweet basil	Ocimum basilicum
	Oregano	Origanum vulgare
Medicinal crops	Aloe	Aloe vera
	Coleus	Solenostemon scutellarioides

***Tab. 3**: Yield Comparisons between hydroponic and open field cultivation [30].*

Type of Crops	**Name of Crops**	**Hydroponic Yield (kg per ha)**	**Open Agriculture Yield (kg per ha)**
Cereals	Rice	13,456.56	841.03-1,009.25
	Maize	8,971.0	1,682.07
	Wheat	5,606.9	672.83
	Oat	3,364.14	953.18
	Soybean	1,682.07	672.83
	Peas	15,699.32	2,242.76
Vegetables	Tomato	403,335.81	11,203.75-22,407.47
	French bean	47,097.96	-
	Beet	22,427.6	10,092.42
	Potato	156,852.29	17,925.98
	Cabbage	20,184.84	14,577.94
	Cauliflower	33,641.4	11,213.8-16,820.7
	Cucumber	31,398.64	7,849.66
	Lady's finger	21,306.22	5,606.9-8,971.04
	Lettuce	23,548.98	10,092.42

1.5.3 Nutrient Solution

All needed nutrients in hydroponics are supplied to the plant through the nutrient solution with the exception of carbon, hydrogen and oxygen, which are carried in the air. Although some inorganic acids are also utilized, the majority of fertilizers used in hydroponics to produce nutrient solutions are highly soluble inorganic salts [31]. Elements absorbed by plant in hydroponics are summarized in Table 4.

***Tab. 4**: Elements absorbed by plants [32].*

Nutrient	Symbol	Forms Absorbed
Nitrogen	N	NO_3^{2-+}, NH_4^{++}
Phosphorus	P	PO_4^{3-}, HPO_4^{2-}, $H_2PO_4^-$
Potassium	K	K^+
Calcium	Ca	Ca^{2+}
Magnesium	Mg	Mg^{2+}
Sulfur	S	SO_4^{2-}
Iron	Fe	Fe^{2+}, Fe^{3+}
Manganese	Mn	Mn^{2+}
Zinc	Zn	Zn^{2+}
Copper	Cu	Cu^{2+}
Molybdenum	Mo	MoO_4^{2-}
Boron	B	$BO_3^{2-}B_4O_7^{2-}$

Some soluble fertilizers used in hydroponics are ammonium nitrate, calcium nitrate, phosphoric acid, nitric acid, etc [31]. These formulations are commercially available in liquid and solid version depending on their growth stage plants can affect the nutrient solution and require different formulations.

Commercially available solutions identify the macronutrient contents as a sequence of N-P-K concentration, expressed in weight percent. For instance various stages of crop and plant development modify the chemical composition of the nutrient solution. Ad example, for lettuce crops an 8-15-16 solution is recommended.

It is crucial to make prompt modifications to the pH, electrical conductivity and water level because these last affect the duration of the solution. The overall volume in the storage tank must remain constant replenishing the water absorbed by the plants and lost through evapotranspiration in order to prevent variations in the nutrient solution. If not the concentration of salts will fluctuate, thus inhibiting the plants capacity to develop properly. Depending on the crop, it is recommended to replace the solution in the tank

every two to three weeks, fully cleaning and disinfecting the tank [33]. Another essential factor to consider is the temperature of the nutrient solution because it affects the capability from of absorb nutrients and yields [34-36].

1.5.4 pH

In addition to the media's physical characteristics, hydroponic crop growth is greatly influenced by the chemical characteristics of the medium, such as pH and electrical conductivity. Depending on the crop type, different pH ranges for soilless media are indicated (see Figure 2). To specify acidity or alkalinity of a nutrient solution is utilized a pH a scale from 1 to 14. Most authors agree that the nutrient solution must have pH between 5 and 7 [31] since it is in such interval that nutrients remain soluble. In addition, optimum pH range of nutrient solution for development of plants in hydroponics was identified in the range from 5.5 to 6.5 [37]. Iron, boron, copper, zinc, and manganese absorption are all inhibited if pH is greater than 7, as the solubility of Fe and $H_2PO_4^-$ diminishes and causes precipitation of Ca and Mg among other chemical interactions between the components of nutrient solution. In addition, the adsorption of nitrogen, phosphorus, potassium, calcium, magnesium, and molybdenum is inhibited if the pH is lower than 5. When pH interval is out of the 5–7, nutrient availability is compromised by chemical reactions that happen between the components of the nutrient solution. Indeed, when some micronutrients, like manganese, are delivered, it sometimes leads to dangerous contamination [33].

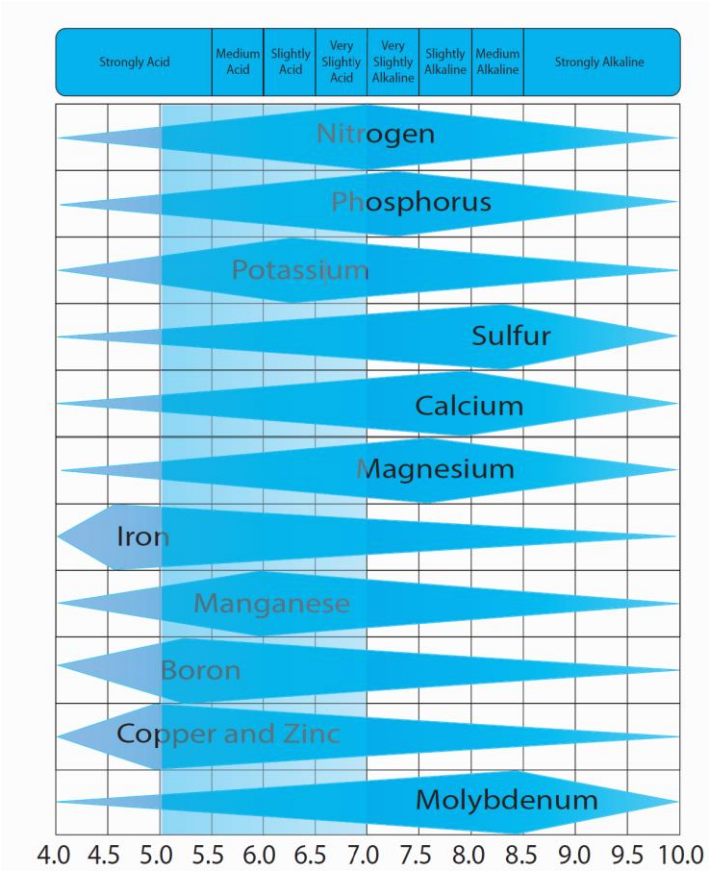


***Fig. 2**: Effect of pH on the availability of nutrients [37].*

1.5.5 Electrical Conductivity (EC)

Electrical conductivity EC is an indicator of the total concentration of ions in a solution. Low EC values represent a lack of nutrients in the form of ions, whereas high values could cause salt stress in the plant [38, 39]. For this reason, EC should be managed within a specified range due to its considerable impact for improving vegetable yield on crop quality and growth. [40, 41]. Optimum range for both EC and pH values for different hydroponic crops is shown in Table 5.

***Tab. 5**: Optimum range of Electrical Conductivity (EC) and pH values for some hydroponic crops [42].*

Crops	EC (dSm^{-1})	pH
Asparagus	1.4 to 1.8	6.0 to 6.8
African Violet	1.2 to 1.5	6.0 to 7.0
Basil	1.0 to 1.6	5.5 to 6.0
Bean	2.0 to 4.0	6.0
Banana	1.8 to 2.2	5.5 to 6.5
Broccoli	2.8 to 3.5	6.0 to 6.8
Cabbage	2.5 to 3.0	6.5 to 7.0
Celery	1.8 to 2.4	6.5
Carnation	2.0 to 3.5	6.0
Courgettes	1.8 to 2.4	6.0
Cucumber	1.7 to 2.0	5.0 to 5.5
Egg plant	2.5 to 3.5	6.0
Ficus	1.6 to 2.4	5.5 to 6.0
Leek	1.4 to 1.8	6.5 to 7.0
Lettuce	1.2 to 1.8	6.0 to 7.0
Pak Choi	1.5 to 2.0	7.0
Peppers	0.8 to 1.8	5.5 to 6.0
Parsley	1.8 to 2.2	6.0 to 6.5
Rhubarb	1.6 to 2.0	5.5 to 6.0
Rose	1.5 to 2.5	5.5 to 6.0
Spinach	1.8 to 2.3	6.0 to 7.0
Strawberry	1.8 to 2.2	6.0
Sage	1.0 to 1.6	5.5 to 6.5
Tomato	2.0 to 4.0	6.0 to 6.5

From the table it is possible to observe that ideal EC range for most of the crops in hydroponics is between 1.5 and 2.5 dS m^{-1}. Osmotic pressure caused by higher EC will prevent nutrients from being absorbed, while lower levels will negatively impact plant health and productivity. However, this parameter does not provide specific information regarding the concentration of each element in the nutrient solution; hence after measuring EC it is essential to add fertilizers in concentration amounts that the plants can absorb (Table 4).

To sum-up, plants respond to various chemical and physical properties and variables in the root zone. In fact the interactions between these factors are complex and much research is needed to optimize the root zone environment. In greenhouse hydroponics major factors used as controlled variables are EC (electrical conductivity) and pH. In plants production is essential keeping all factors within optimum ranges. Indeed, hydroponics grower practices include weekly nutrient analysis to avoid critical accumulation or depletion of some elements in the nutrient solution and adjusting the fertilizer concentration in accordance. Due to the complexity of many component

interactions, optimization has not, however, been entirely completed, and both researchers and commercial practitioners frequently make compromises with the available fertilizer formulations and control strategies.

1.5.6 Water

The increasingly lack of resources makes water more and more important as a resource. In this context hydroponic practices are becoming relevant as a technology for water saving. In addition, consumers are becoming more and more aware of the advantages of choosing high-quality crops cultivated in greenhouses and the demand for hydroponics products is rising in Europe and Asia- Pacific.

Indeed, hydroponics uses less water as compared to the soil farming. In particular, the supplied water is not leached into the soil and it is always available to the plants roots, being constantly in touch among them. Moreover, this procedure uses recovered, filtered, replenished, and recycled water, thus no water is lost. Use of waste nutrient solution as a substitute water supply for hydroponic crop cultivation is possible [43]. From Table 1 is possible to quantify savings aspects in terms of irrigation water, fertilizer and increase in vegetable and water productivity under hydroponic system.

Using hydroponic techniques, in particular NFT based hydroponics, it is possible to reduce irrigation water usage by 70% to 90% by recycling the run-off water. Under controlled hydroponic conditions, it is easy to grow high-quality, high-value vegetables while requiring 85 to 90% less water than conventional soil-based farming. Additionally, salinity, dissolved solids, and pathogens are frequently present in groundwater sources, which can affect plant condition and productivity. Some of these elements may be advantageous to crops, but others should be minimized.

1.6 Environmental factors

The production in a hydroponic greenhouse is influenced by a number of significant environmental parameters, including light, temperature, relative humidity, and CO_2 concentration. The hydroponic greenhouse production is typically negatively impacted by the level of these components (either too low or too high). Thus, it is essential to control them for ensuring a fast and optimal crops growth.

1.6.1 Air temperature

Temperature is an important factor to take into account for plants growth and yield. Indeed, different temperature trends affect chemical reaction and physical properties of plant, in particular plant process as photosynthesis, (the light independent reactions of photosynthesis are dependent on temperature), respiration, and transpiration. For most crops, the highest activity is achieved between 21 °C and 27 °C daytime temperatures in greenhouses [44]. In general, the growth rate of various vegetables and fruits increases as air temperature goes up [44, 46]; as a demonstration, in [47], the biomass provision of the fruit increased with increasing temperature from 18 °C to 25 °C at the same plant. Not only quantitative, but also qualitative characteristics are influenced by temperature: indeed, low temperatures affect directly organoleptic properties of crops. In [48] the impact of temperature variations on agricultural output was examined by Marcelis and Baan Hofman-Eijer, and the results were evaluated versus crop production at constant temperatures. They observed that significant temperature variations between the day and night-time must be prevented because these variations reduce crop quality.

1.6.2 Light intensity

To properly control a confined, environment, artificial light is very important to ensure plant growing and health. It represents an important factor that influences plant growth by affecting photosynthesis, photorespiration, and photoperiodism.

The rate of photosynthesis depends on the availability of different factors as: light, nutrients, water, CO_2, and temperature. However, these transformations of the photosynthetic process requires a lot of energy to occur. Both when the light intensity decreases and is higher, photosynthesis slows down and this affects the plant growth; in the first case low light intensity reduces the leaf carbon assimilation [49], in the second case chloroplasts injury take place.

In photoperiodism, which is the response of plant during the day-night cycle, light role is essential. In fact, along with relative temperature of daylight and dark periods runs different plant responses (e.g. leaf shape, flowering, stem development, etc,).

Light condition depend on seasonal time and weather: in this sense, Winter season become especially a limiting factor. As confirmation of this, for example, in [50] it was shown that treating garlic plants with short days led growth to be inhibited, poor bulb development, and stimulation of secondary growth.

Thus, given temperature condition, the plant growth is further affected by light intensity (irradiance). The response of plant growth to an increase in Photosynthetic photon flux (PPF) is almost linear for many cases (e.g. lettuce) under a controlled environment. External and internal quality of crops are affected both by the lack and by the excess of lighting. One of the biggest issues for the greenhouse sector is represented by light loss since it slows down photosynthesis. Particularly, low photosynthetic photon flux density (PPFD) can contribute to yield and quality limitations [51]. The PPFD measures the amount of the number of photosynthetically active photons (it is the Photosynthetically Active Radiation-PAR) that arrive on a given surface each second; it is measured in micromoles per square meter per second ($\mu mol/m^2/s$). Typically, the photosynthetic photon flux density (PPFD) that is supplied to microgreens for optimal yield, appearance, and nutritional quality in indoor production lies between 300 to 400 mmol m^{-2} s^{-1}.

The linearity in the plant growth rate to daily light integral (DLI, also defined Daily Photosynthetic Photon Flux) is another important index to consider under a controlled environment [52]. The reciprocity between the effects of PPF and photoperiod at the same DLI and the linear response of plant growth to DLI are important in lighting design and in determining photoperiod and PPF to achieve desired DLI.

Regarding the evapotranspiration, it is inversely correlated with leaf stomatal resistance, which is extremely depended on the amount of light reaching the leaves. Also the temperature of leaves and its transfer to the surrounding air is related to the absorption of radiation by leaves. Hence, light intensity on a crop hold is crucial for determining the microclimate in plant grows [53]. Inside the greenhouse crops were subjected to light intensities varying from 100000 lux on clear summer days to 3200 lux on cloudy winter days [54].

The wavelength of light is also important. Plants absorb radiation mostly in the 400-700 nm visible range and, thus, convert CO_2 uptake and water into oxygen and glucose.

To properly control a confined environment, indoor cultivation artificial lightning is increasingly moving towards LEDs (light emitting diodes) usage. LEDs have become central in CEA thanks to their low energy consumption, great wavelength controlling and low heat production. In hydroponic experimental and commercial setup, LEDs are largely employed as light sources [5, 6]. But even more important in recent research is the search for the best combination of the wavelengths of LEDs, affecting yields and promoting plants growth [55, 56]. Thus, the light spectrum is essential because plants have a specific Photosynthetically Active Radiation (PAR) in which they grow and that is defined by

absorption curves of the most important photosynthetic elements: chlorophyll a and b, Phytochrome Pr and Pfr, and carotenoid. The absorption spectra of this elements are shown in Figure 3.

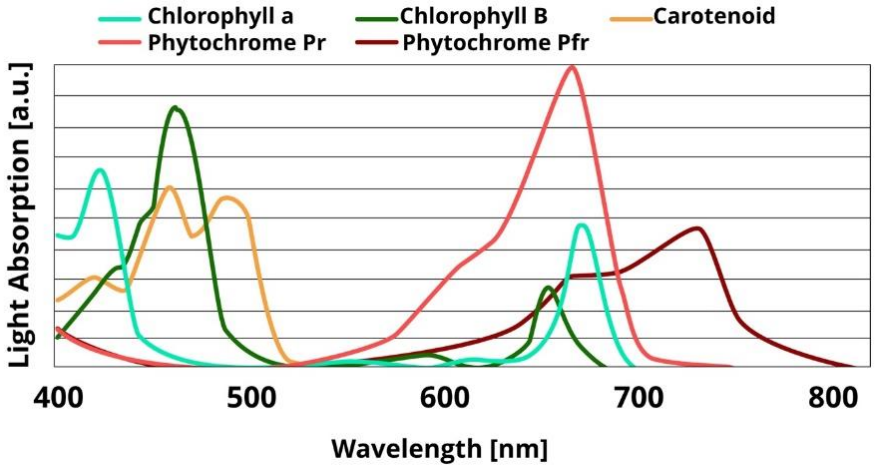


***Fig. 3**: Absorption spectra of most important photosynthetic elements (Figure from https://jhonnydissidence.wordpress.com/tag/absorption-sperctrum/).*

The light quality, which is defined as the color or wavelength that reaches a plant's surface, has a significant impact on plant development [57]. Red (R) and blue (B) lights have the most effects on plant growth because their wavelengths have the greatest impact on photosynthesis due to the absorption peaks of chlorophyll molecules and promoting: the opening of stomata, electron transport, Rubisco activity, antioxidant accumulation, and pigment production [58, 59]. Previous research looked at the activity spectra of higher plants' photosynthesis and it was found out that spectra have action maxima in the B and R ranges [60]. In addition, in controlled environments, the combination of RB LED lights have been shown to be an advanced lighting source for producing several plant species, including lettuce [61-65].

1.6.3 Relative Humidity (RH)

Relative humidity (RH) is a parameter dependent on the temperature and it defines the air water vapor content based on the maximum quantity of water the air can hold at a specific temperature and pressure. It is often expressed as a percentage or ratio of the given water vapor content to the maximum at a given temperature.

In order to maintain a high standard of plants, it is necessary to regulate the relative humidity inside the greenhouse. For most of the crop the standard relative humidity is 60-75 %. In closed greenhouses, the most damaging high humidity occurs in the few hours following sunrise because solar radiation that is transferred into the interior speeds plant

transpiration [66]. But too high relative humidity is also dangerous for plants because it promotes the germination of pathogenic spores [67].

Although relative humidity is frequently used to evaluate environmental humidity, it does not directly indicate what induces transpiration and evaporation. However, the driving force is measured by the vapor pressure deficit (VPD), which means that the rates of transpiration and evaporation are proportional to the VPD. VPD can be defined as the difference between the quantity of moisture in the air and the amount it can hold when it is saturated at the same air temperature. The SI unit of VPD is kPa (kilopascal), or kg m^3.

As a consequence, the maximum water vapor pressure increases as the temperature increases because air can store more water vapor at higher temperatures. If VDP is too low, transpiration will be inhibited leading toa possible condensation on leaves and surfaces inside controlled cultivation environment.

In a soilless cultivation environment, the ideal range for VPD is from 0.8 to 0.95 kPa, with an optimal setting of around 0.85 kPa. To measure humidity variations there are humidity sensors that use electronic devices based on changes in electrical capacitance or resistance.

1.6.4 CO_2 Concentration

CO_2 concentration is an environmental factor that with small changes in concentration can significantly impact on the rate of photosynthesis of plants. Plants that grow in controlled environments with CO_2 enrichment improve biomass and the generation of secondary metabolites by raising net photosynthetic rates [68]. Many studies have been done on effects of CO_2 enrichment and it was concluded that dry weight of plant, plant height, number of plant leaves, and lateral branches of plant increases with the increase in CO_2 enrichment in greenhouse; this happens increasing the rate at which carbon is incorporated into carbohydrate in the light-independent reaction [69]. Moreover, also temperature and light factors can increase with the elevated CO_2 concentrations [69].

On the other hand, if plants are actively photosynthesizing and the culture room is not actively ventilated during the photoperiod, if there is no CO_2 enrichment and no human activity, CO_2 concentration can drop to a very low level, close to the CO_2 compensation point. The degree of fluctuation in CO_2 concentration inside the culture room depends on the volume of the culture room (buffering capacity), human occupancy, and plant activity. It is often difficult to detect low level CO_2 in controlled environment because plants grow slowly without any symptoms under low CO_2 concentration [7].

2 Experimental tests: Monitoring Study of a Standard Hydroponic cultivation system

2.1 Experimental evaluation of a standard hydroponic greenhouse

A microclimatic monitoring was carried out in a standard hydroponic greenhouse as a comparison with the monitoring tests done in the controlled environment agriculture object of this PhD thesis. The acquired data were elaborated to evaluate also the trend of the microclimatic parameters inside the hydroponic greenhouse and to identify the best optimization asset.

2.1.1 Description of the experimental environment

The standard cultivation environment is represented by an operating hydroponic greenhouse of 500 m^2 (10 m x 50 m) (Width x Depth). This greenhouse (Fig. 4a, b) is located at 850 meters above sea-level in Vinchiaturo (Molise region- Italy).

a **b**

***Fig. 4**: Hydroponic greenhouse structure pictures; **a**) external environment; **b**) indoor cultivation environment.*

The span greenhouse structure is made of metal and polycarbonate with side openings made of polyethylene film, stretched over the structure by means of a winding roller system. Side openings are provided with anti-aphid meshes. There are two entrances at opposite ends of the building consisting of doors 3 m x 3 m. The height of the greenhouse is 6.50 m at the ridge line and 3.40 m at the eave line.

It is a closed cycle cultivation system since it allows the water and nutrients dissolved in it to be recovered and reused several times in the same production cycle. Along the side

there are two cultivation tanks connected to each other in Deep Water Culture hydroponic system: the plants are raised on floating supports placed in tanks filled with water and nutrient solution. Water inside tanks is lightly moved by individual pumps: this is necessary both for oxygenation and for recycling of the water. The size of the tanks is 4 m x 46 m and the internal content is water and nutrient solution dissolved in it. The water contained inside one tank is 60 m^3 for a total of 120 m^3 of water in both tanks. The tanks are insulated with EPS, i.e. highly layered synthesised expanded polyester with a thickness of 10 cm' placed both along the walls and on the floor. The insulation of the tanks prevents temperature fluctuations of the water' avoiding unfavourable temperatures to plant growth.

A system of nozzles is placed above the tanks to moisturize canopies if necessary.

This hydroponic greenhouse is very basic and due to that it is not equipped with artificial lamps or LEDs to enhance crops growing. Main crops cultivated in this hydroponic greenhouse were lettuce (*Lactuca sativa*, variety Salanova), aromatic herbs, and edible flowers. These were raised in polystyrene raft (33 cm x 55 cm x 5.5 cm) and were pushed from the bottom side to the main entrance for the final harvest to allow the transplantation of new plants. Each plant's hole on the raft had a 'x'-shaped slit at the base that allowed the roots to pass. As the roots grew longer' they dipped into the nutrient solution to absorb more nutrients.

Microclimatic measurements were carried out on two complete crops growth cycles of lettuce, monitoring, in particular, two targeted floating rafts with 4 plants each (Figure 5). These last were positioned in such a way as to promote the lettuce head to grow and have no roots competition for nutrients.

Fig. 5*: Arrangement of lettuce inside the polystyrene raft.*

Tests followed the ongoing management of the greenhouse: in fact, the first monitored test was on 28th March - 27th April, 2022 and was characterized by the absence of a mechanical ventilation system; the second test occurred on 14th July - 13th August, 2022, using one mechanical fan installed in May 2022. This fan (Figure 6) is 1 m x 1 m in size, with an air extraction capacity of 17000 m^3/h, equal to 7.6 forced air changes per hour. The fan was fixed close to the main entrance to the greenhouse and was activated when inner temperature reached 30 ± 3 °C. However, it did not have any automatic control for the on/off cycles based on the humidity inside the hydroponic greenhouse, which, in general, could reach higher humidity levels (~ 90 %).

Fig. 6*: Fan near the main greenhouse entrance.*

2.1.2 Experimental procedures

For the experimental tests the inner environment was divided in different zones both on the aisle and on the selected cultivation side tank. In particular, the selected cultivation tank was divided in four zones (Z1, Z2, Z3, Z4) to identify different growth stages, while the central aisle in three sections (C1, C2, C3) (Figure 7).

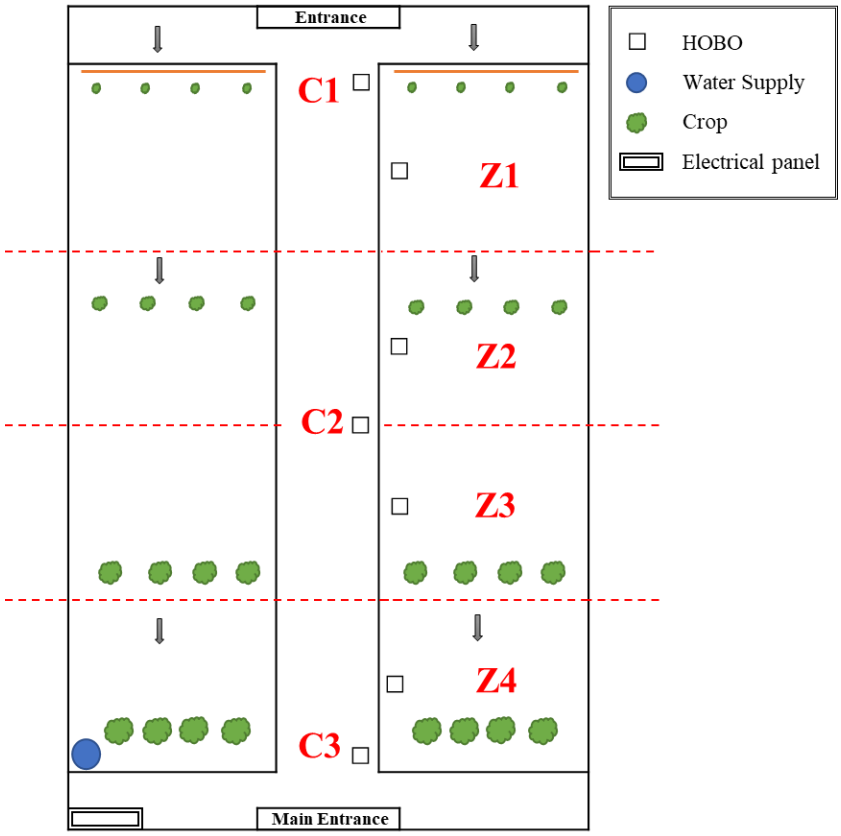


Fig. 7*: Scheme of the cultivation environment. The blue circle represents the water source tank, the green shapes the crops, the arrow the direction of the path followed by the plant; Z1, Z2, Z3, and Z4 indicate the four stages of the cultivation tank; C1, C2, and C3 are the area of the central aisle close to the entrance, in the middle, and close to the other entrance.*

Seven HOBO sensors were used for Temperature, Relative Humidity, and Light intensity monitoring. They were placed close to both greenhouse entrances and close to the selected rafts of leafy crops for the monitoring of growth phases. Four of them were equally distributed at a distance of 30 cm from the cultivation bed and other three in the central aisle at a hight of 1.80 meters (Figure 8a, b). The following Table 6 summarizes the main features of HOBO sensor.

***Tab. 6**: HOBO Sensors Specifications.*

Measurements	**Accuracy**
Temperature	
Measurement range: -20 °C to 70 °C (-4 °F to 158°F)	
Response time: 6 minutes (to 90 % in airflow of 1 m/s)	± 0.35 °C from 0 °C to 50 °C (0.63 °F from 32 °F to 122 °F)
Relative Humidity (RH)	
Measurement range: 5 % to 95 % RH	±2.5 % typical, 3.5 % maximum, from 10 to 90 % RH
Response time: 1 minute (to 90% in airflow of 1 m/s)	
Light Intensity	
Range: 1 to 3000 footcandles (lumens/ft^2) typical 0-32.300 lumens/m^2 [0-3,000 footcandles (lumens/ft^2)]	
External Input	
External sensors for temperature, AC current, AC voltage, CO_2, 4-20 mA, 0-10 VDC	±2 mV, ±2.5 % of absolute reading
Input range: 0 to 2.5 VDC	
Output power: 2.5 VDC at 2 mA, active only during measurements	

a **b**

***Fig. 8**: Example of HOBO sensors fixed in C3 (a) and in Z2, near the raft of leafy crops (b).*

The growing cycle of Salanova multi-leaf lettuce was followed during the experimental monitoring using HOBOware Pro supervision system that measured and historicized data for 30 days (day-night cycle). The sampling time for each parameter was of 6 minutes.

From an agronomic point of view, parameters such as root length, height of the head part, diameter, number of leaves, and final plant weight was monitored. In particular, root length was considered because its apparatus is essential for the absorption of nutrients. Initially, measurements of the agronomic parameters were carried out by completely

removing the plant from the tray, but in this way the plant suffered a great deal of stress due to the manipulation, as can be seen from the graph in Figure 9, which represents the root growth of the lettuce.

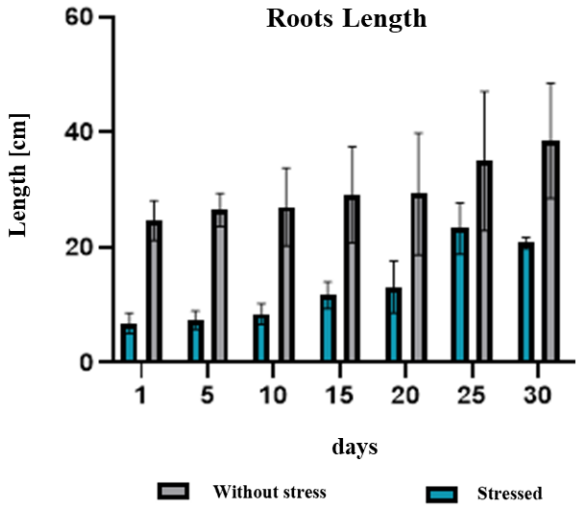
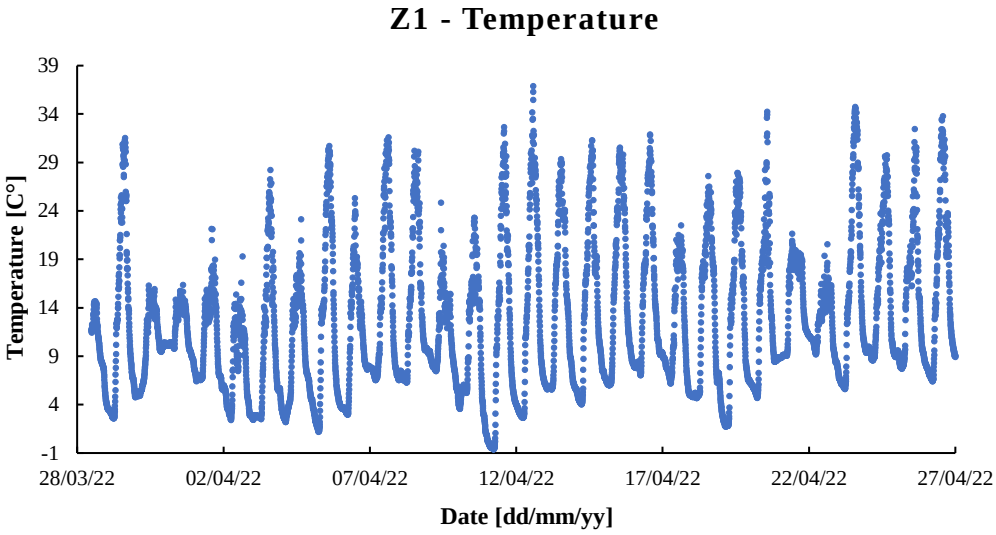


***Fig. 9**: Length of lettuce roots without stress and with stress conditions.*

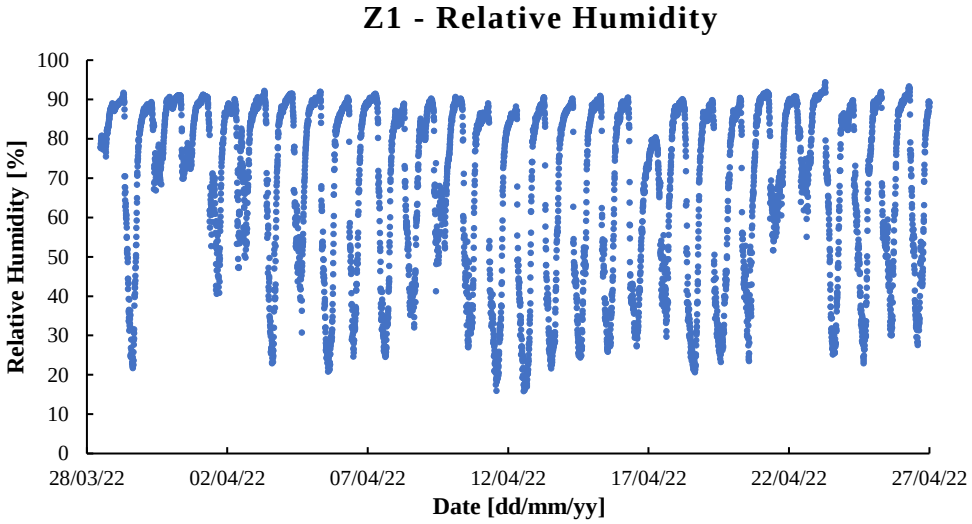
Comparing the roots with those of lettuces from other trays that were at the same stage of growth, it was observed that the roots were more developed both in quantity of roots emitted and in relation to length. For this reason, subsequent measurements were made leaving the plant undisturbed, that is, not extracting it completely from the trays.

2.1.3 Results and Discussion

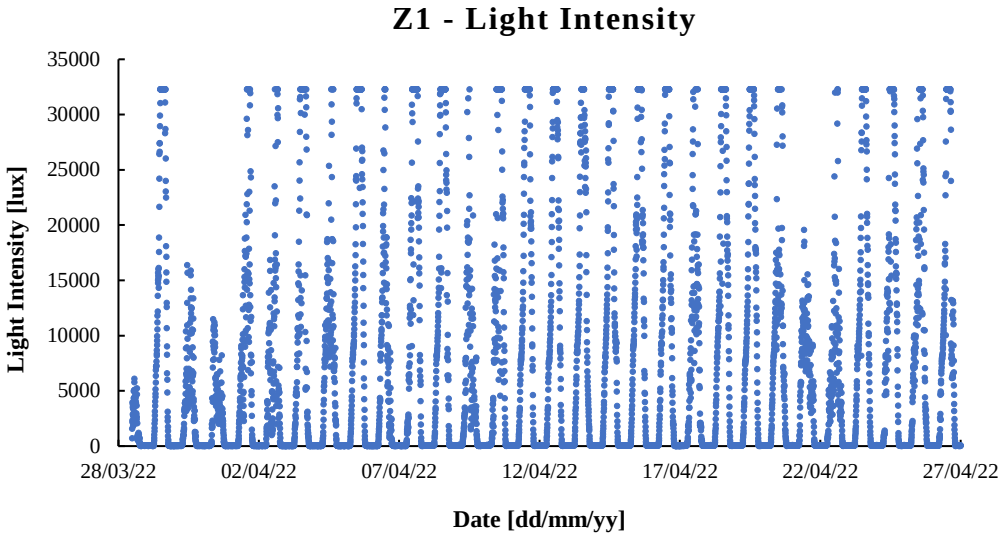
First monitoring test of the cultivation cycle (from March 28th to April 27th) was characterized by unfavourable weather conditions. Trends in temperature, relative humidity and light intensity showed no substantial differences between the different areas, for both Z and C. Indeed, the mean value of $\Delta T_{x,y} = T_x - T_y$ with $x, y = C1, C2, C3, Z1, Z2, Z3, Z4$ is always less than 1.5 °C in absolute value. Analogously, the mean value of $\Delta \mathrm{RH}_{x,y} = RH_x - \mathrm{RH}_y$ is less than 5% in absolute value. Thus, the trends of the variables are independent of the measurement area and, as an example, the Z1 area was chosen (Figure 10a, b, c).


Z1 - Temperature
Temperature [C°]
39
34
29
24
19
14
9
4
-1
28/03/22
02/04/22
07/04/22
12/04/22
17/04/22
22/04/22
27/04/22
Date [dd/mm/yy]

a


Z1 - Relative Humidity
Relative Humidity [%]
100
90
80
70
60
50
40
30
20
10
0
28/03/22
02/04/22
07/04/22
12/04/22
17/04/22
22/04/22
27/04/22
Date [dd/mm/yy]

b

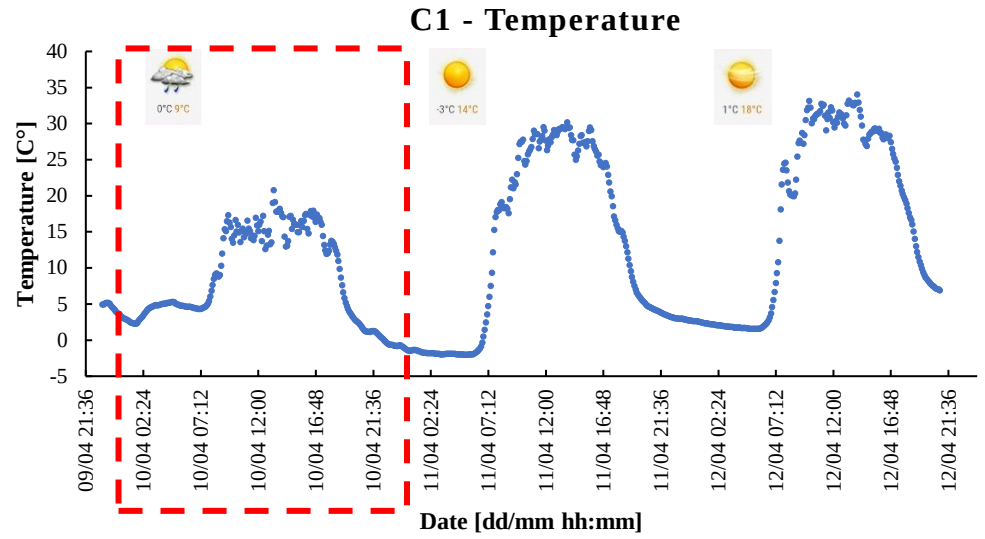


c

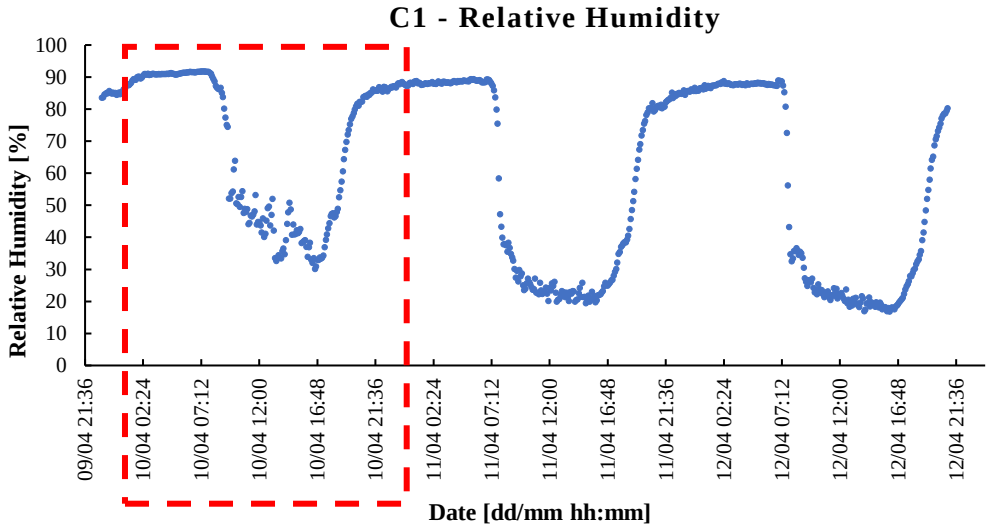
***Fig. 10**: Growing cycle in Z1 (March 28th - April 27th, 2022) temperature (**a**) , relative humidity (**b**) and light intensity (**c**) trend.*

In particular, the first cycle was cold and rainy, explaining the thermal excursion of 22.33 °C with a standard deviation of 6.63 °C. This happened also because of the absence of mechanical ventilation and not proper insulation of the structure that affected the internal cultivation environment.

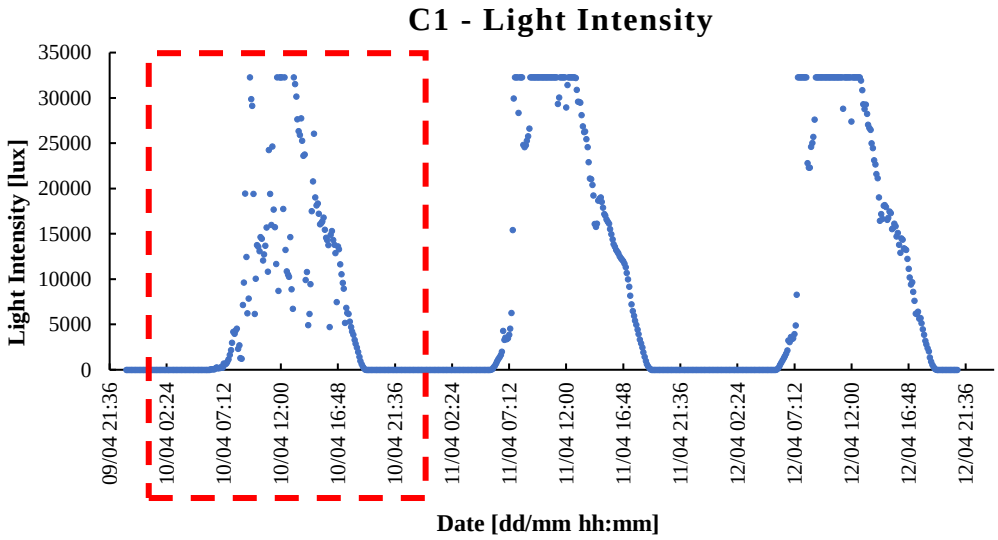
Likewise, the relative humidity was strongly affected by the bad weather: the daily variation average of the relative humidity was 57.63 %, with a standard deviation of 15.91 %. The light intensity (measured in lux) was representative of the external weather conditions and was almost similar in all the zones; the average of maximum values achieved 30000 lux, optimal value for plants growing (the optimal range in indoor systems is 20000-50000 lux). The strong variability of temperature and humidity can be better understood analysing data during a short time period of meteorological instability, showed in Figures 11a, b, c.


C1 - Temperature
Temperature [C°]
40
35
30
25
20
15
10
5
0
-5
09/04 21:36
10/04 02:24
10/04 07:12
10/04 12:00
10/04 16:48
10/04 21:36
11/04 02:24
11/04 07:12
11/04 12:00
11/04 16:48
11/04 21:36
12/04 02:24
12/04 07:12
12/04 12:00
12/04 16:48
12/04 21:36
Date [dd/mm hh:mm]

a


C1 - Relative Humidity
Relative Humidity [%]
100
90
80
70
60
50
40
30
20
10
0
09/04 21:36
10/04 02:24
10/04 07:12
10/04 12:00
10/04 16:48
10/04 21:36
11/04 02:24
11/04 07:12
11/04 12:00
11/04 16:48
11/04 21:36
12/04 02:24
12/04 07:12
12/04 12:00
12/04 16:48
12/04 21:36
Date [dd/mm hh:mm]

b


C1 - Light Intensity
Light Intensity [lux]
35000
30000
25000
20000
15000
10000
5000
0
09/04 21:36
10/04 02:24
10/04 07:12
10/04 12:00
10/04 16:48
10/04 21:36
11/04 02:24
11/04 07:12
11/04 12:00
11/04 16:48
11/04 21:36
12/04 02:24
12/04 07:12
12/04 12:00
12/04 16:48
12/04 21:36
Date [dd/mm hh:mm]

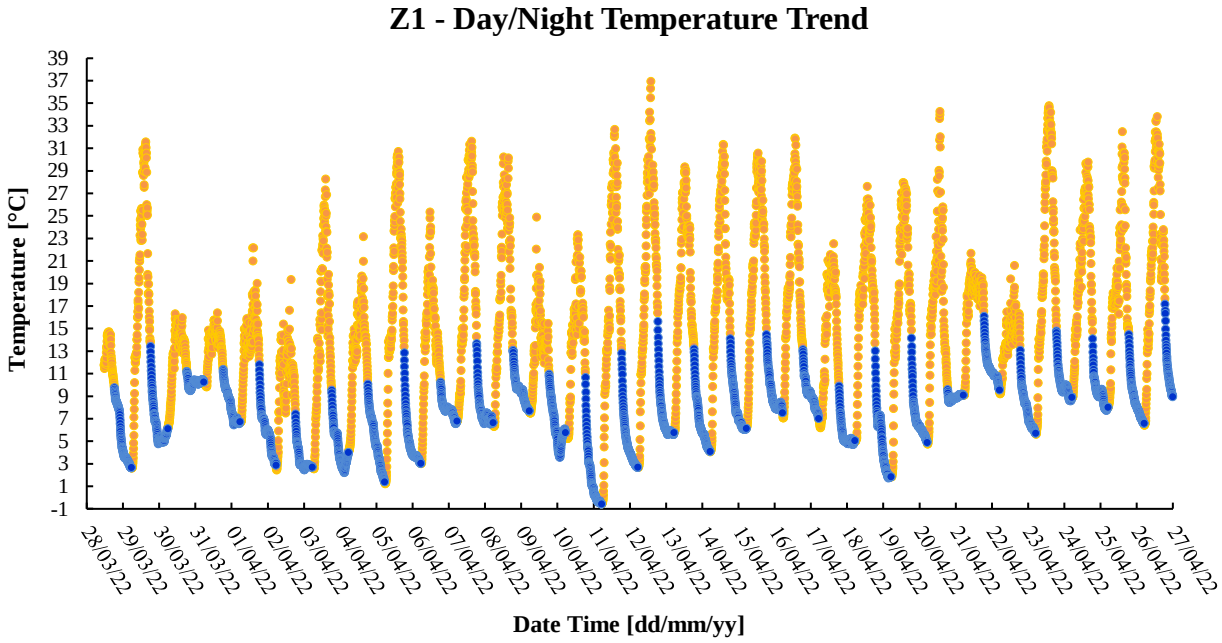
c

***Fig. 11**: Few day data (April 10th - April 12th, 2022) of total cultivation growing cycle in C1, C2, C3 (March 28th - April 27th, 2022): **a.** Temperature trend; **b.** Relative Humidity trend; **c.** Light Intensity trend.*

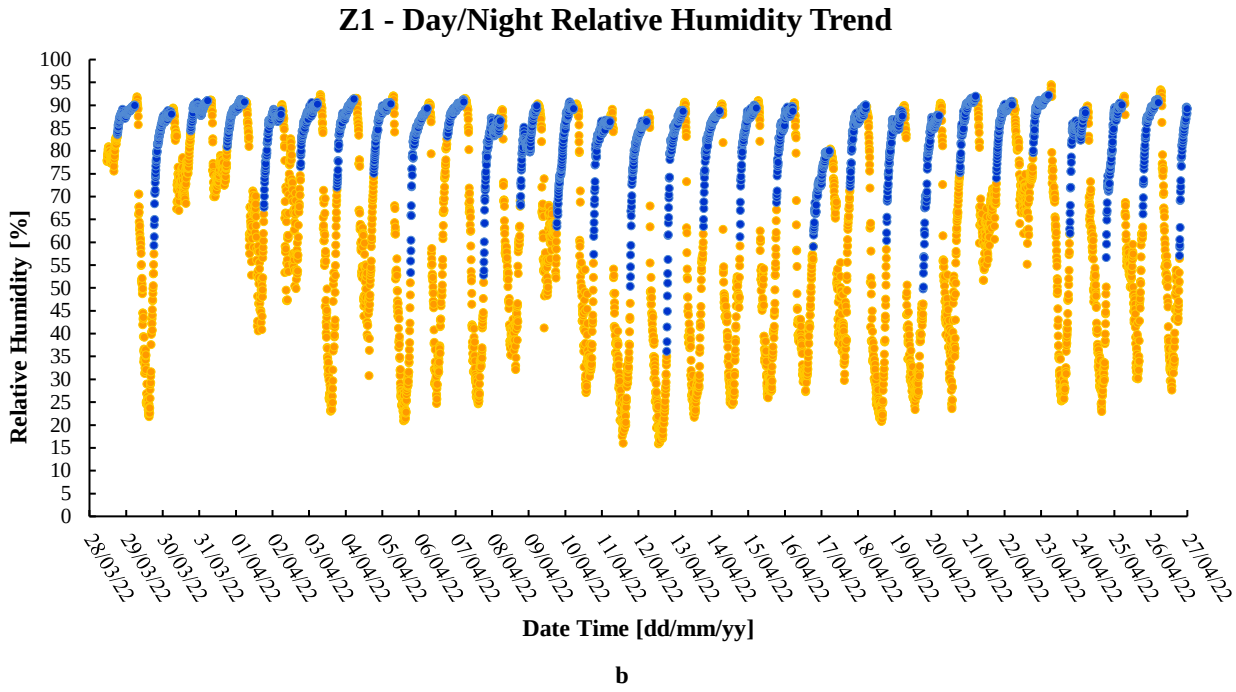
In fact, during the raining days like the April 10th, very clear and strong fluctuations of temperature and relative humidity were evident and strictly related to the outdoor environment. For example, the indoor day temperature had an average value of 15.83 °C on April 10th, while on April 11th and 12th it was 26.95 °C and 30.11 °C.

Thereby, on April 10th relative humidity increased of 20 % and the light intensity decreased of 20.49 % compared to the following days. There is a clear necessity for a well-insulated structure and an automatic ventilation system to minimize temperature fluctuations.

A cultivation environment's unfavourable conditions can be observed not only through daily fluctuations but also through the day/night temperature and relative humidity trend. These are more aptly illustrated in the Figures 12a, b, where the previous concept of data similarity is applicable.



a



b

Fig. 12*: Day/night trend of the first test:* ***a****) Temperature;* ***b****) Relative Humidity.*

In the cultivation environment, the maximum temperature reached during the first test was 36.91 °C while the minimum was -0.59 °C. In addition, a maximum relative humidity of 94.47 % and a minimum relative humidity of 88.25 % was acquired.

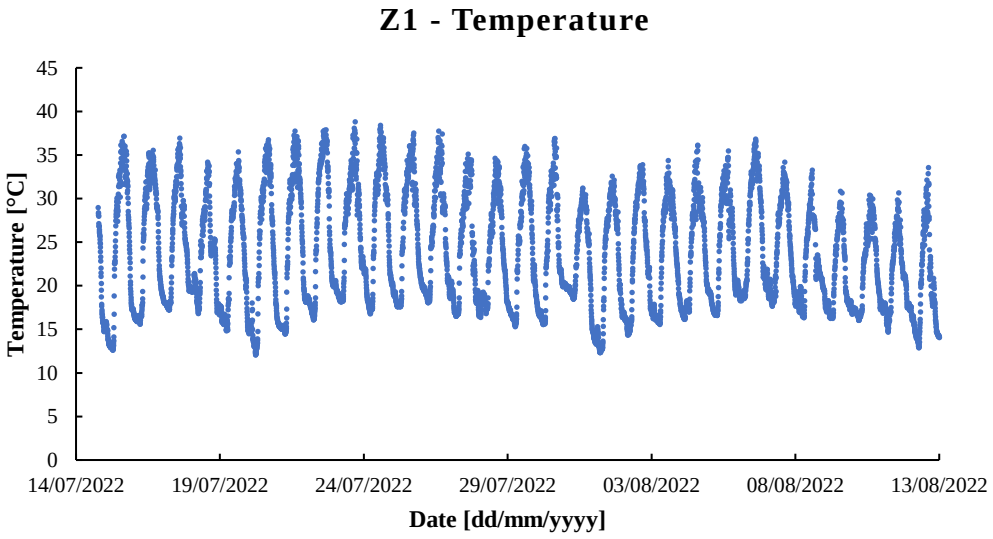
The average daily temperature during the test was 16.96 °C with a standard deviation of 3.84 °C, while the average of the night temperature was 7.06 °C with a standard deviation of 2.36 °C. The average daily relative humidity was of 56.28 %, with a standard deviation of 4.49 %, while the average of the night relative humidity was 84.59 %, with a standard deviation of 1.35 %. It is possible to observe that at lower temperature values correspond to higher relative humidity levels, for example from March 30th to April 1st. This general behaviour in the cultivation environment can increase the risk of having a slower and more difficult plant growth; in the present case this caused physiopathies, especially rots as *Sclerotinia sclerotium* and fungal disease as *Bremia lactucae* (Figures 13a, b).

a **b**

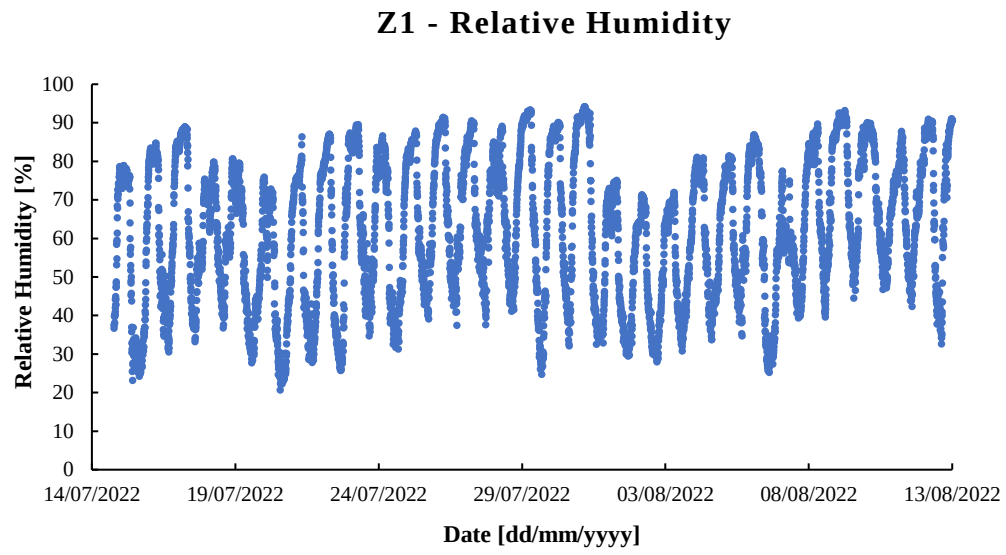
***Fig. 13**: **a)** Sclerotinia sclerotium, and **b)** Bremia lactucae on lettuce plant.*

From July 14th to August 13th, 2022, a second monitoring test of the cultivation cycle was conducted. A mechanical fan was continuously regulated during the monitoring test. The average daily temperature in the side cultivation zones and in the central aisle decreased of about 15 % compared to the first test, and of about 6 % in the central aisle, despite maximum temperature values achieved across the greenhouse of 34 °C with an average of 22 °C.

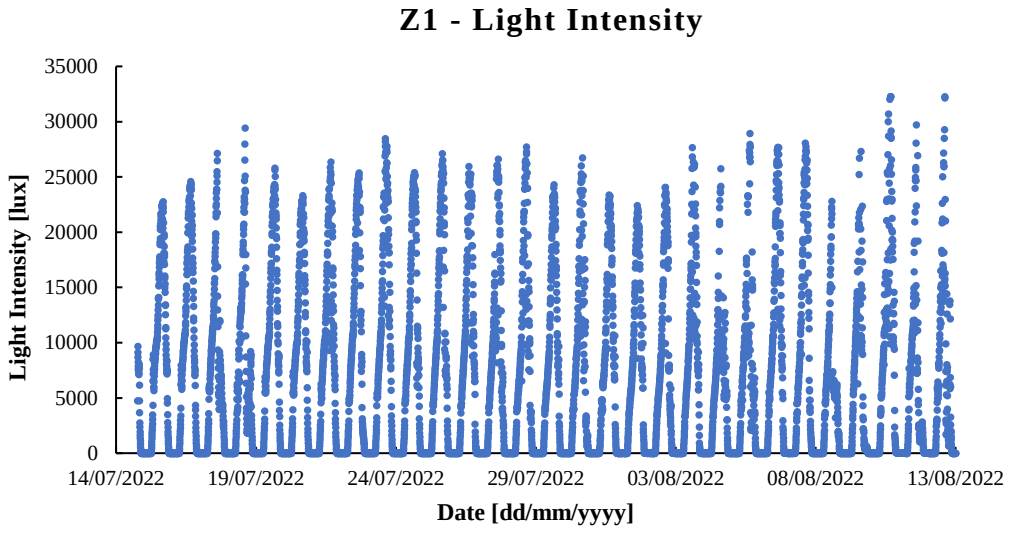
As for the first test, also trends in temperature, relative humidity and light intensity of the second test showed no particular variations between Z and C areas being the mean value of $\Delta T_{x,y}$ always less than 1.5 °C in absolute value and of $\Delta \mathrm{RH}_{x,y}$ less than 5% in absolute value. Thus, Z1 trends are shown below in Figures 14a, b, c, as an exemplification case of area Z.



a



b

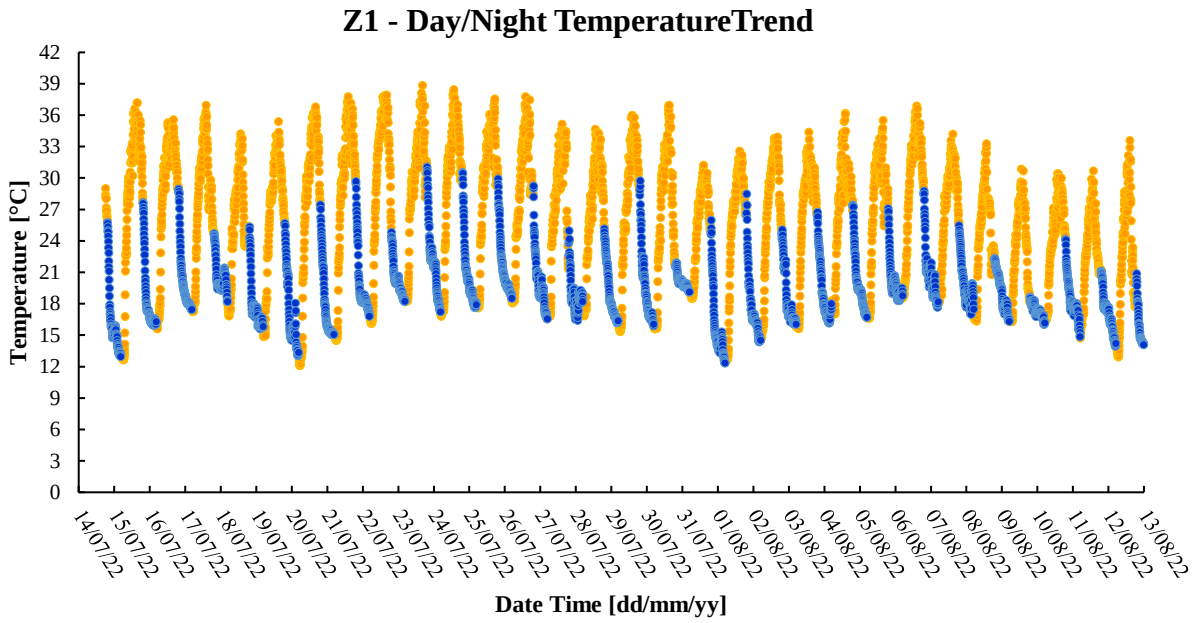


c

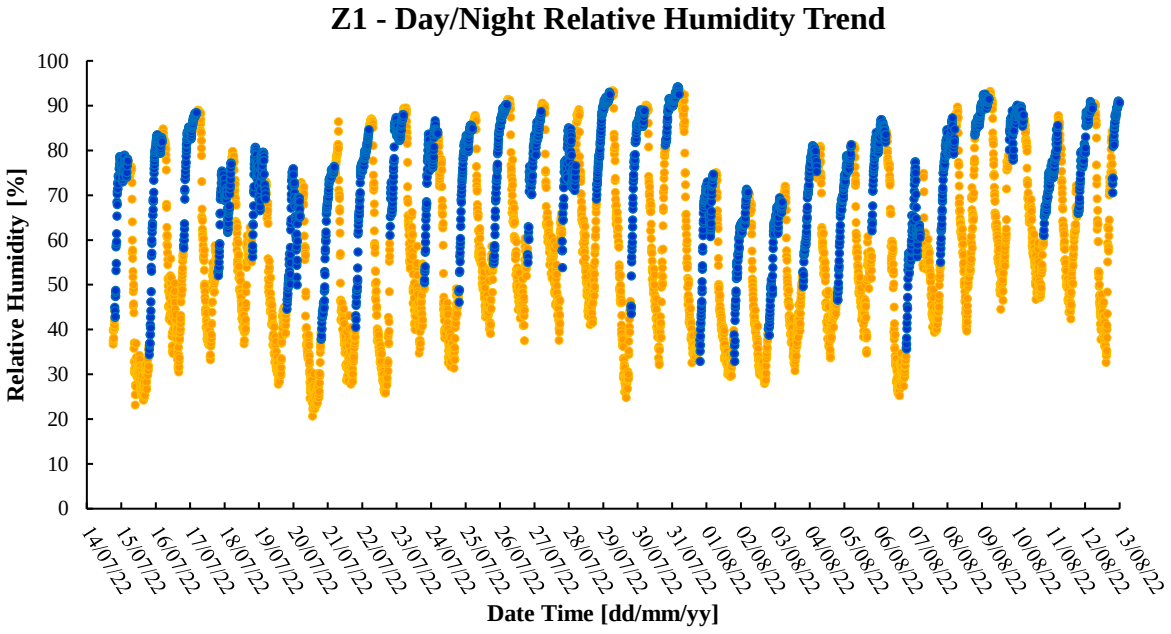
***Fig. 14**: Growing cycle in Z1 (July 14th - August 13th, 2022) temperature (**a**) , relative humidity (**b**) and light intensity (**c**) trend.*

Compared to the first test, the second showed a decrease of the daily average temperature in the cultivation and central areas of approximately 3 °C: in fact, it decreased from 21.88 °C to 19.07 °C in Z areas and from 23.17 °C to 20.16 °C in C areas. The relative humidity dropped by 11.86 % above the side cultivation bed (Z) and by 12.56 % in the central aisle (C). It is clear that there was a uniform decrease in relative humidity as compared to the Z areas since the range in average daily relative humidity fell from 57.05 % to 51 % in the Z areas and from 58.54 % to 52.01 % in C areas.

From the day/night temperature and relative humidity trends shown in Figures 15a, b, it is possible to discuss about measurements influenced by the use of the fan.



a



b

Fig. 15*: Day/night trend of the second test:* ***a****) Temperature;* ***b****) Relative humidity.*

The maximum temperature reached in the cultivation environment during the second test was 38.81 °C, while the minimum was 12.07 °C. The maximum relative humidity acquired was of 94.21 % and the minimum relative humidity was of 20.68 %. Nevertheless, these maximum values not suitable for plants, in this case the average daily

temperature was 26.70 °C with a standard deviation of 6.29 °C, while the average of the night temperature was 18.85 °C with a standard deviation of 3.19 °C. The average daily relative humidity was of 54.42 %, with a standard deviation of 18.09 %, while the average of the night relative humidity was 75.28 %, with a standard deviation of 12.55 %.

Despite the considerable temperature and relative humidity fluctuations in the two tests, there were no physiopathies in the second one because of the higher temperature, lower relative humidity and reduced fan-induced fluctuations.

It is evident from this study that a ventilation system operated by a set of stable probes will improve this hydroponics greenhouse's management consistently, reducing day-night excursion and improving the production efficiency. Considering that cultivated plants were growing vegetatively, it is indicated to turn on the fans whenever the relative humidity is above 80% and the temperature has been consistently above the set-point of 27 °C. This prompt action would prevent the temperature from dropping too low on rainy days and during the winter/spring season, especially in case of the not well insulated structure of the greenhouse.

In both first and second test there were no substantial differences regarding the agronomic parameters measurement, for this reason the following Table 7 shows values chosen from the second monitoring test to be brief.

Tab. 7: A*gronomic parameters measurement*s on lettuce seedlings.

Date	zone	ID Plant	Roots Length (cm)	Head Length (cm)	Diameter (cm)	leaves n°	Final Head Plant Weight (g)
14/07/2022	Z1	1	4	4.5	5	12	25
14/07/2022	Z1	2	4	4	7	14	28
14/07/2022	Z1	3	4	4.4	4.5	12	32
14/07/2022	Z1	4	4	4	5.5	12	26
04/08/2022	Z2	1	38 (+4)	8	20	48	/
04/08/2022	Z2	2	40 (+4)	9	12	40	/
04/08/2022	Z2	3	24 (+4)	5	18	43	/
04/08/2022	Z2	4	35 (+4)	8	17	52	/
13/08/2022	Z3	1	60 (+4)	12	21	100	180
13/08/2022	Z3	2	45 (+4)	12	19	80	230
13/08/2022	Z3	3	50 (+4)	11	21	97	220
13/08/2022	Z3	4	48 (+4)	12	21	120	200

In this table values stop at zone Z3 because plants were harvested in the boundary part between Z3 and Z4, as in this area a good percentage of lettuce plants are already to be removed from the cultivation tank.

It possible to underline that, lettuce cultivated according to the planting pattern of four seedlings per tray, had a yield that meets production standards; indeed, in a growing cycle, that lasted an average of 30 days, lettuce plants as a whole reached a weight of about 200 g, this making them suitable for sale.

3 Experimental tests: Monitoring Study of a Small Scale Hydroponic cultivation system

3.1 Microclimatic parameters characterization in a confined agricultural environment for the supply of the air conditioning

3.1.1 General Context

The study of a small scale hydroponic system was the main objective of the work. The experimental set up was realised in in the university experimental laboratory, that fully deal with the concept of CEA.

The adaptation of the environment agriculture condition to plant needs to improve agriculture efficiency is possible studying the management of the CEA, enhancing, in particular, the importance of microclimatic control [70, 71]. Depending on its location, CEA designs and environmental control techniques can differ significantly [72]. CEA systems are distinguished by a clear separation between the controlled agriculture environment (indoor) and the external, uncontrolled environment (outdoor), even if the outdoor characteristics are taken in account to better regulate the inside environment and dampen the climatic variations of the outdoors during the day and seasons [73]. As a result, in controlled agricultural environments with microclimate management, outdoor environmental variability has no impact on plant cultivation, and indoor microclimatic parameters can be independently managed, increasing the harvest efficiency. It is evident that in CEA, optimal monitoring of the indoor microclimate becomes fundamental to enhancing yield and promoting the growth and health of plants [74, 75]. Indeed, as a result of the interaction between the environment and the plant, an unusual variation in the microclimate can induce both morphological and physiological alterations in plants [76]. In this context, it becomes essential to consider appropriate control systems and locations inside the confined agricultural environment as well as the monitoring and management of environmental factors [77]. The main controlling factors for the microclimate in agricultural environments are, in particular, lighting and air conditioning, through which it is possible to work directly and indirectly on both quantitative and qualitative parameters of crops [78].

Thus, before the realization of the small scale hydroponic system, a preliminary study to evaluate the distribution of the microclimatic parameters in the confined agricultural environment conditioned was necessary to be carried out.

The air conditioning system was handled by ensuring the microclimatic parameters of a CEA for successive hydroponic lettuce plants cultivation. A microclimatic control unit, which simulates the plant, was used to achieve this goal. The measures of temperature, humidity, globe thermometer temperature, and air speed, were performed at different fan speeds for air conditioning supplying with a perforated duct.

3.1.2 Description of the experimental environment

Tests for microclimatic parameters characterization were conducted in the closed environment represented by the university experimental laboratory. It has an inner volume of 40 m^3 and dimensions of 4.1 m x 3.6 m x 2.7 m (Width x Depth x Height) (Figure 16). Sandwich panels made of 4.0 cm polyurethane foam were used to insulate the environment on the roof and the outside walls.

A handling unit of 500 m^3/h provide the conditioned air supplied by a perforated duct of 0.25 m in diameter and 3 m in length, with a 5 series of circular holes of different size. A recovery grid is located in the corner on the opposite side of the supply duct, at a height of about 60 cm.

The relative humidity of the environment was ensured by the work of an immersed electrode steam humidifier.

It was possible to regulate the handling unit and the humidifier to stabilize the indoor microclimatic parameters measuring the recovery temperature T_R (probe installed on the recovery side of the handling unit) and the room humidity U_{CTR} (combined T-RH probe located at the centre of the environment).

A supervision system allowed the data collection and historicization thanks to a set of probes and a microclimatic control unit that was used instead of lettuce plants.

3.1.3 Experimental procedures

The microclimatic control unit was placed at nine different locations equally distributed into the controlled agricultural environment (see Figure 16), for a uniform characterization of microclimatic parameters of the closed environment.

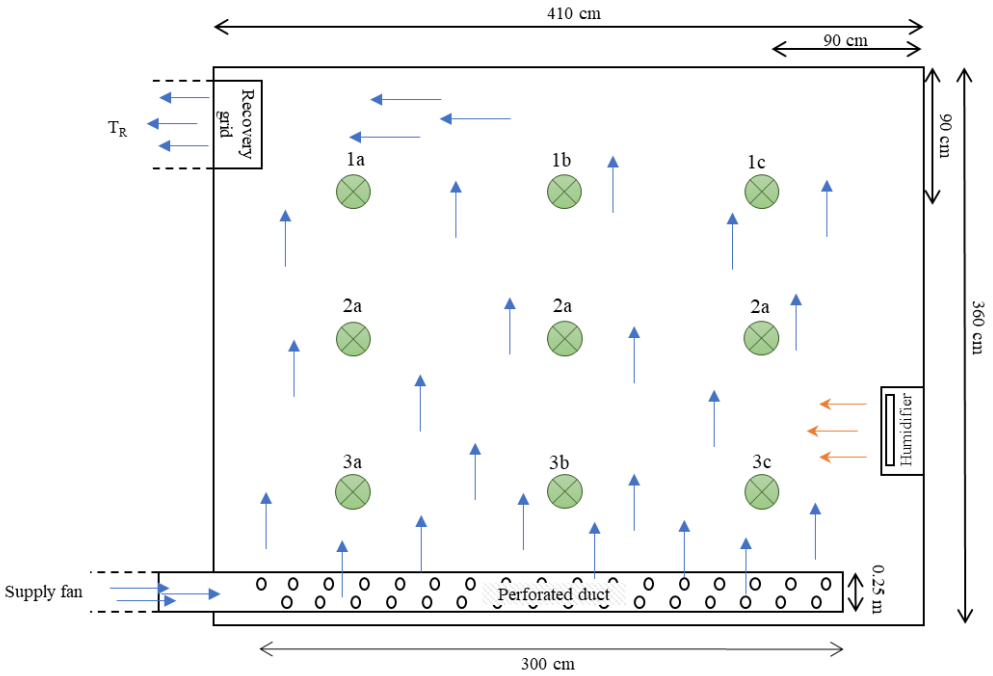


***Fig. 16**: Spatial locations in the controlled agricultural environment, equipped with a supply fan, a perforated duct, a recovery grid, and a humidifier.*

A set of probes were fixed at a height of 1.05 m for the acquisition of the main microclimatic dimension (temperature, radiant temperature, humidity, and air speed) and, simulating the height of a shelf for the positioning of plants.

The probes used for the measurement tests are from the brand Centraline by Honeywell, except for the globe thermometer which is LSI LASTEM. Table 8 shows the characteristics of the probes used.

***Tab. 8**: Probes.*

Probes	Accuracy
Humidity and Temperature Room Sensors, range 5–95% -30 − 70 °C, output 0–10 Vcc	±0.2 K at 25 °C ±3 % at 25 °C 30–70 % rh ±5 % at 25 °C 10–30 % 70–90 % rh ±10 % at 25 °C 5–10 % 90–95 % rh
Globe thermometric probe, range -30 – 70 °C	0.15 °C
Duct humidity-temperature sensor, range 5–95% 30–70°C, output 0–10 Vcc	±0.3 K at 25 °C ±2.5 % at 20 °C 10–95 % rh
Hot wire anemometer, range 0–20 m/s, -50 +50 °C	<10 %

For an optimum growth of lettuce crop it was chosen on one hand an environment set-point temperature of 21 ± 1 °C, as constant day temperature; on the other the indoor humidity was fixed at 70 % ± 5 %. The fan (ebm-papst K3G 190, with M3G055 – BI

electronic motor) was set at three different airflow speeds: 30 %, 50 %, and 80 % of its maximum speed.

All of the data characterizing the indoor microclimate for each of the nine space positions were collected once the system reached the regime. The acquisition time was of 5 minutes, that corresponded to enough time to gain a thorough knowledge of the local conditions. The sampling time for each parameter was set to 5 seconds.

When data acquisition was completed, the collected data were transferred in spreadsheet format onto a personal computer.

3.1.4 Results and Discussion

To investigate the distributions of the physical parameters inside the controlled environment, the gathered data were elaborated. These estimates are made on the valid assumption that the environment's physical conditions are nearly constant at regime. Thus, it is possible to state that the temporal mean of all the data gathered by each probe is a realistic estimation of the measured parameters and the standard deviation can be taken into account as its uncertainty. The main microclimatic parameters measured at 30 %, 50 %, and 80 % of fan speed, respectively, are shown in Tables 9, 10, and 11.

***Tab. 9**: : Average values and standard deviation of measured parameters at 30 % airflow speed.*

		1a	1b	1c	2a	2b	2c	3a	3b	3c
T_{CTR} [°C]	$\bar{x}$	24.66	25.39	25.00	24.51	25.27	24.83	25.45	25.11	24.68
	σ	0.05	0.06	0.04	0.04	0.06	0.04	0.05	0.03	0.04
U_{CTR} [%]	$\bar{x}$	73.28	67.41	66.46	71.29	71.04	69.17	65.05	67.62	73.71
	σ	2.87	1.44	2.01	4.27	1.94	0.13	1.63	3.31	0.52
T_{cr} [°C]	$\bar{x}$	23.80	24.27	24.00	23.69	24.11	23.78	24.30	24.13	23.95
	σ	0.03	0.05	0.04	0.08	0.03	0.04	0.06	0.07	0.05
U_{cr} [%]	$\bar{x}$	77.34	70.43	70.55	79.39	73.09	71.98	71.00	77.08	76.07
	σ	2.28	0.76	0.69	3.42	1.20	0.39	0.53	3.25	0.37
T_S [°C]	$\bar{x}$	37.98	38.59	38.87	37.15	38.25	37.89	38.05	38.91	38.08
	σ	0.08	0.19	0.15	4.84	0.12	0.38	0.26	0.14	0.14
U_S [%]	$\bar{x}$	19.48	19.13	18.57	20.36	19.13	19.20	18.84	18.84	19.64
	σ	0.11	0.08	0.07	0.53	0.10	0.33	0.05	0.21	0.15
T_G [°C]	$\bar{x}$	20.90	21.48	21.20	20.18	21.44	20.95	21.52	21.17	22.93
	σ	0.09	0.15	0.07	0.08	0.09	0.05	0.09	0.14	0.08
$\dot{V}_{ren}$	$\bar{x}$	130.34	127.65	130.34	127.60	127.72	131.09	128.70	130.74	128.82
	σ	0.82	1.08	1.53	1.46	2.04	1.58	1.36	2.21	2.41

***Tab. 10**: Average values and standard deviation of measured parameters at 50 % airflow speed.*

		1a	1b	1c	2a	2b	2c	3a	3b	3c
T_{CTR} [°C]	$\bar{x}$	22.36	23.61	23.79	23.55	23.62	24.46	23.34	22.85	23.38
	σ	0.14	0.10	0.05	0.08	0.12	0.05	0.06	0.09	0.14

U_{CTR} [%]	$\bar{x}$	62.78	70.56	68.92	68.86	72.11	68.21	70.63	70.37	69.91
	σ	0.32	0.70	0.41	0.69	0.93	0.32	1.49	0.52	1.04
T_{cr} [°C]	$\bar{x}$	21.70	22.92	23.05	22.89	22.93	23.73	22.71	22.43	22.72
	σ	0.11	0.04	0.05	0.03	0.09	0.05	0.08	0.05	0.10
U_{cr} [%]	$\bar{x}$	65.20	72.31	71.48	71.94	75.10	72.35	71.09	72.43	74.07
	σ	0.40	1.04	0.11	1.56	1.36	0.69	1.80	0.49	1.80
T_S [°C]	$\bar{x}$	31.97	32.90	33.98	31.93	32.50	33.49	32.41	31.97	33.35
	σ	0.09	0.25	0.80	0.21	0.12	0.62	0.15	0.09	0.30
U_S [%]	$\bar{x}$	21.62	22.67	21.79	24.67	22.71	21.36	24.32	23.65	21.73
	σ	0.12	0.20	0.98	0.38	0.26	0.24	0.14	0.10	0.31
T_G [°C]	$\bar{x}$	21.98	21.95	22.27	21.90	22.27	22.75	21.64	20.75	21.76
	σ	0.14	0.33	0.03	0.14	0.54	0.04	0.17	0.19	0.30
$\dot{V}_{ren}$	$\bar{x}$	223.98	231.93	222.30	230.25	233.10	229.05	230.62	229.05	234.58
	σ	1.66	3.54	9.15	1.45	3.18	2.92	2.69	3.41	2.76

***Tab. 11**: Average values and standard deviation of measured parameters at 80 % airflow speed.*

		1a	1b	1c	2a	2b	2c	3a	3b	3c
T_{CTR} [°C]	$\bar{x}$	21.90	22.01	22.07	22.08	21.77	21.83	21.13	21.70	21.85
	σ	0.07	0.08	0.05	0.08	0.08	0.04	0.14	0.14	0.11
U_{CTR} [%]	$\bar{x}$	69.09	71.19	70.62	69.91	67.67	68.12	62.63	63.48	65.99
	σ	0.26	0.29	2.19	0.81	0.59	0.51	0.64	0.73	0.63
T_{cr} [°C]	$\bar{x}$	21.81	21.90	21.66	21.93	21.43	21.40	20.32	21.03	21.46
	σ	0.03	0.08	0.05	0.05	0.05	0.07	0.10	0.06	0.05
U_{cr} [%]	$\bar{x}$	70.50	71.89	72.32	71.52	70.19	71.14	67.12	67.15	67.62
	σ	0.07	0.40	1.69	0.63	0.65	0.74	0.32	0.30	0.32
T_S [°C]	$\bar{x}$	29.68	29.96	29.14	29.36	29.06	28.87	28.57	29.03	28.68
	σ	0.74	0.45	0.30	0.10	0.23	0.21	0.06	0.35	0.07
U_S [%]	$\bar{x}$	28.32	27.56	27.96	27.87	28.03	28.37	27.35	27.82	28.94
	σ	0.36	0.16	0.16	0.09	0.15	0.24	0.24	0.41	0.16
T_G [°C]	$\bar{x}$	20.19	20.65	20.41	21.03	20.39	19.81	20.70	20.25	20.33
	σ	0.07	0.19	0.12	0.30	0.20	0.18	0.07	0.14	0.10
$\dot{V}_{ren}$	$\bar{x}$	392.87	391.67	392.46	391.11	394.24	394.10	388.96	394.01	386.98
	σ	15.12	6.09	3.98	2.97	4.15	2.99	2.67	6.16	4.80

The air conditions of the supply air (T_s and U_s), close to the crop (T_{cr} and U_{cr}), and in the middle of the environment (T_{CTR} and U_{CTR}) were also gathered in addition to the renewal air ($\dot{V}_{ren}$). Additionally, a globe thermometer's radiant temperature (T_G) was gathered to complete the characterization of the indoor microclimate.

At a fan speed of 30 % and at mean renewal air flow $\dot{V}_{ren}$ of 129.28 m^3/h (Table 9), the spatial mean of crop temperature T_{cr} in the environment was 24.03 °C and the spatial mean of crop relative humidity U_{cr} was 74.10 % with a mean supply air temperature T_s of 38.20 °C. At a fan speed to 50 % the mean $\dot{V}_{ren}$ increased to 229.43 m^3/h (Table 10). In this configuration, there was a decrease of mean T_s to 32.72 °C with a consequent decrease of the mean T_{cr} to 22.79 °C. The U_{cr} reached a mean value of 71.77 %.

Similarly, as Table 11 shows, $\dot{V}_{ren}$ increased at 391.82 m^3/h at a fan speed of 80 %, while the mean supply temperature dropped further to T_s 29.15 °C, and T_{cr} fell to 21.44 °C. In these conditions, the mean U_{cr} is 69.94 %.

In order to fully determine whether this environment is suitable for growing plants like lettuce, it is also essential to consider the effect of the temperature by thermal radiation. The globe thermometric temperature T_G, which is the temperature recorded by the Vernon globe thermometer, was measured to evaluate this effect. The spatial mean of T_G varied with air flow rate and was, respectively, 21.31 °C, 21.92 °C, and 20.43 °C for 30 %, 50 %, and 80 % airflow rate. It was evident from comparing this variation to that of T_{CR} that thermal radiation and air flow rates had a significant effect on plants, local temperature. In order to provide a term of comparison for each acquisition, a room temperature T_{CTR} was acquired concurrently with these measurements using a temperature probe set on the room's center pedestal. The use of the perforated duct in such an application could guarantee the best environmental conditions for plant growth, as mentioned by La Fianza et al., 2019.

It is possible to observe that the mean crop temperature was about 3.0 °C above the set point temperature at fan speed of 30 %. A too-low air flow rate was the main cause of this. Indeed, a lower air flowrate requires a greater supply temperature, leading to a higher indoor temperature. It is important to highlight that the recovery temperature measurement probe is installed on the handling unit's recovery side in order to better understand this point (Figure 1). This temperature had a value of 21 °C ± 1 °C in each test. The average temperature difference between the crop and set point was 1.8 °C at 50 % fan speed and 1.4 °C at 80 %. This indicates that the best temperature management was made possible by the fan's increased speed, which ensures roughly 10 air exchanges per hour.

In regards to the relative humidity, it should be highlighted that the higher values were obtained along the diagonal, going from the humidifier towards the return grid, at a fan speed of 30 %. At 50 %, the same trend can be seen, whereas at 80 %, a more uniform distribution is maintained. This indicates that a suitable mixing of water vapor inside the environment can be achieved by setting the fan speed at 80 %.

From all test, the mean air speed in close proximity to the crop is around 0.1 m/s, which is a very good value to avoid plant stress from transpiration.

Although these are only preliminary results, determining how the main microclimatic parameters are distributed indoors is important to being able to control the conditioning systems while there are crops present.

3.2 Experimental evaluation of a mini hydroponic cultivation system

After the preliminary investigation of microclimatic parameters distribution, the aim of the work was the realization and study of a hydroponic cultivation system in scale, completely confined.

3.2.1 Description of the experimental environment

The experimental cultivation system was placed in the university experimental laboratory, properly set up for the purpose. The laboratory consisted of a climatized controlled environment and was arranged as shown in the following Figure 17.

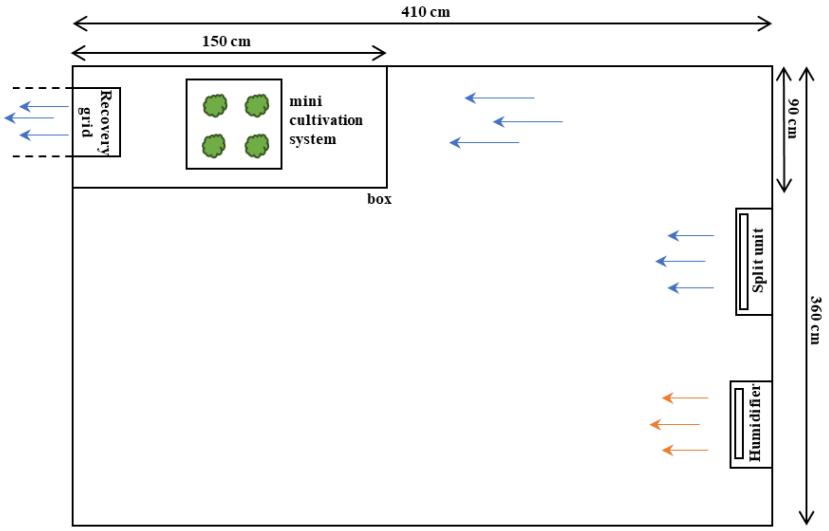


***Fig. 17**: Controlled agricultural environment, equipped with a split unit, a humidifier, a recovery grid, and a metal box with a mini cultivation system inside.*

The climatic room requires a basic-level control. Indeed, the confined environment reproduced the temperature conditions using indoor split unit (Figure 19a) with an inverter heat pump (Figure 18), and a humidifier (Figure 19b).

***Fig. 18**: Heat pump external units.*

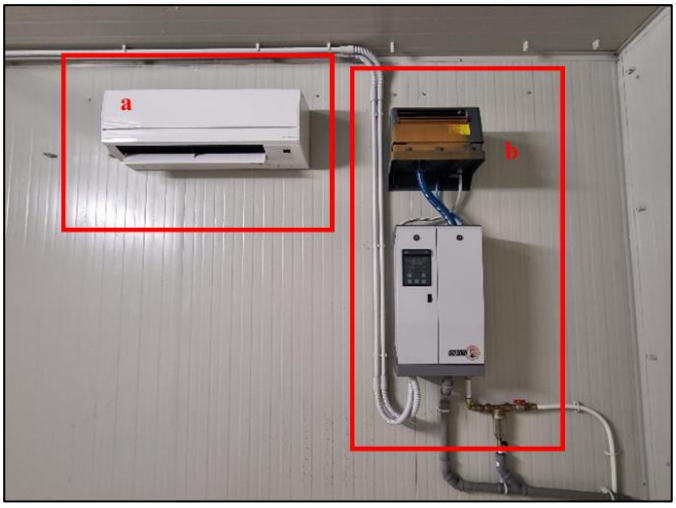
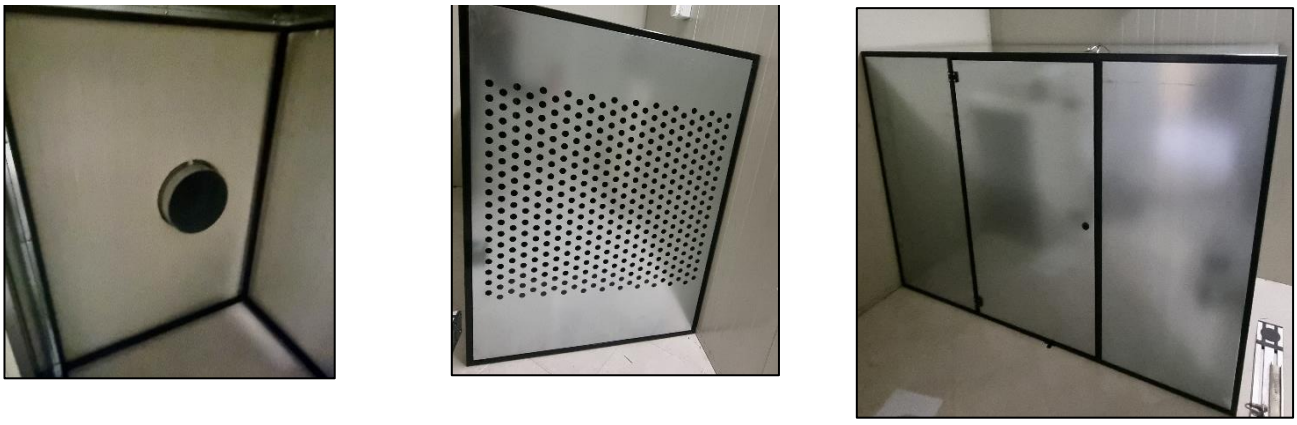


***Fig. 19**: **a**) Split unit, **b**) On/off immersed electrodes humidifier in the confined environment.*

The air exchange per hour in the environment was of about 330 m^3 h^{-1}. The steam for relative humidity conditions were produced with an on/off immersed electrodes humidifier. The on/off cycles are managed through a probe installed in the confined environment. The steam rate is set with the on board control panel at a value in the range 0.9 – 3.0 kg/h.

3.2.2 Description of the small scale hydroponic cultivation system

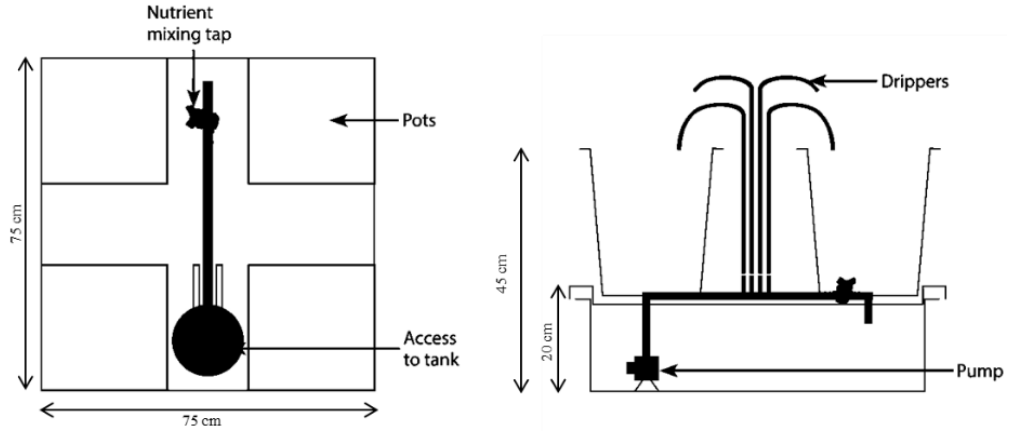
It consisted of an external metal box, placed in the climatized controlled environment in the corner side close to the recovery grid. The box had a dimension of 170 cm x 90 cm x 120 cm (Width x Depth x Height) and was perforated on the opposite side of the recovery grid (Figures 20a, b, c).



a **b** **c**

***Fig. 20**: **a**) side of the box at the recovery grid, **b**) perforated side for air inlet on the opposite side of the recovery grid, **c)** outside of the box.*

The small hydroponic cultivation system (75 cm x 75 cm x 40 cm ,Width x Depth x Height) was placed inside the box. This plant, shown in Figures 21a-b-c-d-e, consists of a nutrient solution tank, a support tray with 4 pots, 4 drippers, a submersible pump.



a

b **c** **d**

e

***Fig. 21**: **a**) Design of the hydroponic cultivation system; **b**) enlarged clay pebbles; **c**) dripper; **d**) nutrient solution in the tank; **e**) complete hydroponic plant.*

The tank, made of plastic, has a capacity of 50 litres and its black color permits to avoid the growth of algae. It contains the nutrient solution that is moved from the tank towards the four drippers into the pots. The solution consisted of water mixed with nutrients from Mills Nutrients brand, in particular "Basis A&B" for vegetative growth (Nutrients composition: Nitrogen (N): 3.2 %, Phosphate (P_2O_5): 4.1 %, Potassium (K_2O): 2.7 %, Magnesium (MgO): 0.7 %, Calcium (CaO): 4.9 %, Sulphur (SO_3): 1.4 %, Boron (B): 0.0045 %, Copper (Cu): 0.0025 %, Iron (Fe): 0.037 %, Manganese (Mn): 0.0200 %, Molybdenum (Mo): 0.0006 %, Zinc (Zn): 0.0100 %). A pH^- solution was used for pH regulation. The monitoring of the nutrient solution was possible thanks to a pH/Temperature-meter and a EC-meter. The acidity was maintained within 5.6-6 pH, while the Electrical Conductivity at 1.08 dS/m.

The circulation of the solution is possible thanks to a submersible pump, suitable for 24/7 hours use (Figure 22). This pump has dimensions of 14.0 cm x 10.2 cm x 11.0 cm and a capacity of 1000 L/U; the maximum head is of 160 cm and the minimum water depth is 2.5 cm; the power consumption is 22 Watt.

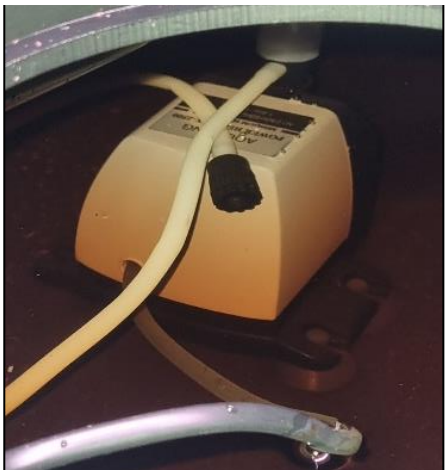


***Fig. 22**: Low water multifunctional immersible pump.*

All plastic pots, with a capacity of 11 litres each, were filled with enlarged clay as growing medium. The enlarged clay increased the oxygenation and the moisture retention capacity around root area. A lettuce of the Canasta cultivar was planted for each pot; lettuce was chosen not only as a comparison with the study conducted in the standard hydroponic greenhouse, but also because it is often used as a model crop in studying plant responses to LED lighting environment in indoor factory with artificial lighting, due to its fast growth, short production period, low energy demand, and high nutritional values [79].

Thus, a LEDs bar of 1 meter was installed above the hydroponic cultivation system as light source (Figure 23).

***Fig. 23:** Small hydroponic system illuminated by the LED bar.*

To evaluate the minimum height h at which the light cone would illuminate the entire cultivation area, a simple calculation is performed. The system shape is a square with a side $l = 75$ cm. The LEDs bar was placed on its diagonal $D = \sqrt{2}l \approx 106$ cm. To

evaluate h, it is needed to underline that the orthogonal cross-section of the light cone is well represented by a isosceles triangle with basis equal to D and opposite angle $\alpha = 120°$. Therefore, the following relation is valid:

$$h = \frac{D}{2}\cot\frac{\alpha}{2} \approx 31 \text{ cm}$$

This is the minimum height, but, in order to leave space for the plants and the operator, the LED was placed at 65 cm from the system. This is showed in Figure 24.

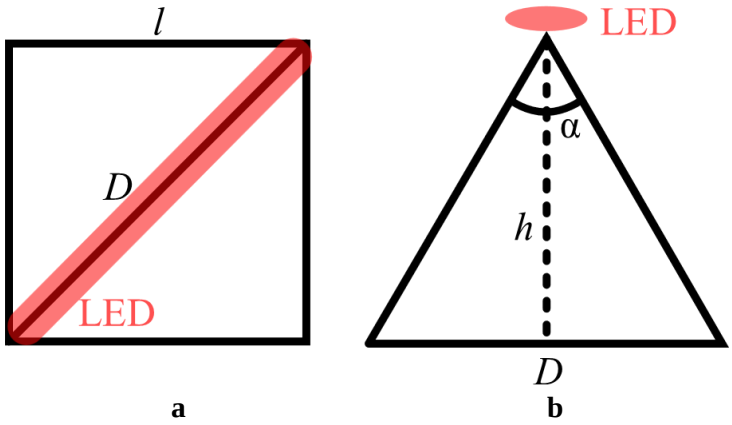


***Fig. 24**: **a)** Schematic view of the system seen from above and **b)** orthogonal section of the light cone, where D is the diagonal of the LED, l is the length of the system side, α is the angle of the light cone, h is the height of the light cone.*

The LEDs bar used was the Flora LED SLIM 35W by Domotrick with a power supply (Figure 25a, b). It consisted of aluminium body extruded and then anodized, equipped with transparent UV resistant polycarbonate cover screen. The multi LED boards have latest generation 5630 LEDs mounted on special low voltage electronic circuit (24vdc). These full spectrum LEDs not only utilise the Blue and Red spectrum part of conventional light sources, but also contain a UV spectrum part that further stimulates growth. The 6500 °K LED adds a further blue component to the total spectrum, in this way it stimulates more and more plants growing (Figure 26). Its technical specifications are briefly as follows:

- Working voltage: 24Vdc (230Vac via external driver).
- Power factor: 96%.
- Total power: 25 W
- LED type: Osram Duris E5 6500°K - Domotrick Full Spectrum.
- Optic: 120°.
- Cooling: Passive.
- Operating temperatures: 0 - 45 °C.

- Total PPF (1 meter version - 30W): 250 µmols/sec at 20 cm - 70 µmols/sec at 40 cm.
- PPF Efficiency/Watt: 2.2 µmols/Watt.
- Total PPFD: 722 µmols/m^2/sec.
- Emission spectrum: White 6500 °K + Full Spectrum.
- Light coverage: over 200 cm x 60 cm.

a

b

***Fig. 25**: **a**) Flora LED SLIM 35W by Domotrick, **b**) Mean Well LPV-100-24 driver LED.*

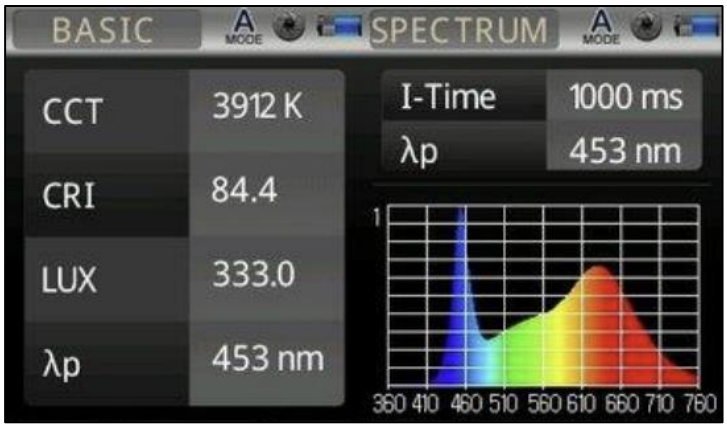


***Fig. 26**: LED bar total spectrum (6500 °K + Full spectrum). In particular, CRI: Color Rendering Index; CCT: Correlated Colour Temperature; λp: Peak Wavelength.*

Thanks to a Plug-In Timer (Figure 27) it was possible to set a light cycle of 16/8 hours (day/night: switching on the LED bar at 6:00 a.m. and switching off at 10:00 p.m.) for the lettuce cultivation.

***Fig. 27**: Plug-in Timer.*

3.2.3 Experimental procedures

For the experimental tests a set of probes were set to acquire data. The control of all the devices and component installed at the laboratory was made thanks to a set of probes and a supervisory system (COACH AX of CentraLine by Honeywell).

In particular, four HOBO sensor were installed inside the box, at each pot close to the lettuce seedlings for the monitoring of Temperature, Relative Humidity (respectively, $T_{\_IN}$ and $U_{\_IN}$), and Light intensity to measure PAR, as shown in Figure 28.

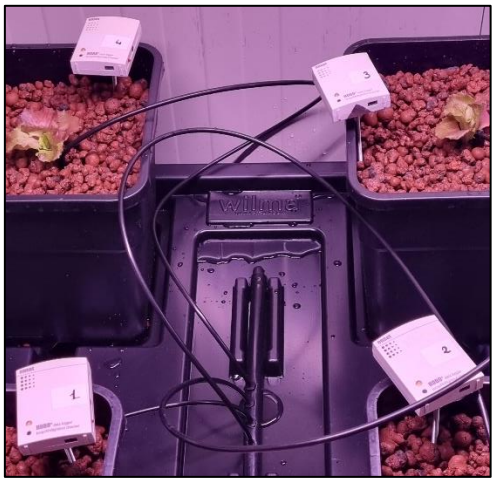


***Fig. 28**: HOBO sensor in the pots.*

Outside the box, at air inlet holes, the probe used for the measurement tests of temperature and relative humidity ($T_{\_OUT}$ and $U_{\_OUT}$) was from Centraline by Honeywell (for details see Table 1) and for CO_2 measure (CO_{2_IN}) from Carel S.p.A. (output. 0-10 Vcc; accuracy: + 2 %) (Figure 29). Another CO_2 probe was installed in the recovery grid to measure carbon dioxide (CO_{2_OUT}) at the exit of the box.

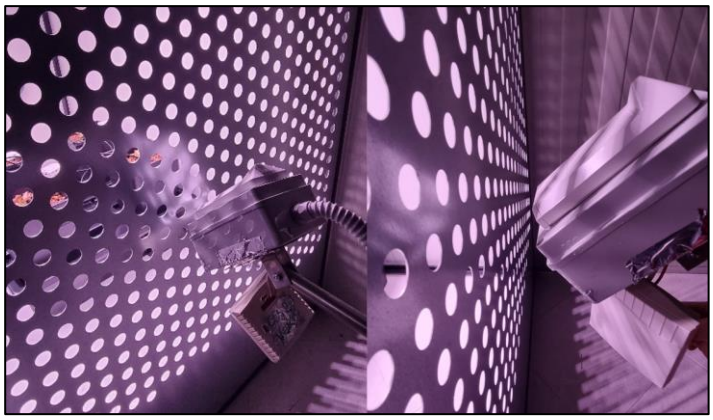


***Fig. 29**: Probes at the ventilation probes.*

Four lettuce plants were cultivated and their development was monitored during one production cycle of 30 days (23rd December, 2021 - 21st January, 2022).

Measurement variables acquisition began when the boundary conditions were stable for at least 15 minutes. A constant daytime indoor temperature of 21 °C, with a dead band of ± 2 °C and a relative humidity set-point of 65 %, with a dead band of ± 5 %, were fixed for optimal lettuce growth and regulated by the sensor probe at air inlet holes. Sampling time for each parameter was of 4 minutes for inbox probes and of 20 minutes for probes outside the box.

Lettuce seedlings were selected randomly from a commercial nursery tray and they were transplanted once seedlings reached the stage of 3 real leaves (after 22 days from sowing in traditional farming). Nutrient solution was renewed after 15 days and the sequent nutrient mix dosages were used for 50 litres of water, as shown by Table 12.

***Tab. 12**: Nutrient mix dosage of new solution.*

	WEEK 1	**WEEK 2**	**WEEK 3**	**WEEK 4**
BASIS A	90 ml	105 ml	140 ml	170 ml
BASIS B	90 ml	105 ml	140 ml	170 ml
pH		5.6 - 6		
EC	1.0–1.2	1.2–1.4	1.4–1.6	1.6–1.8

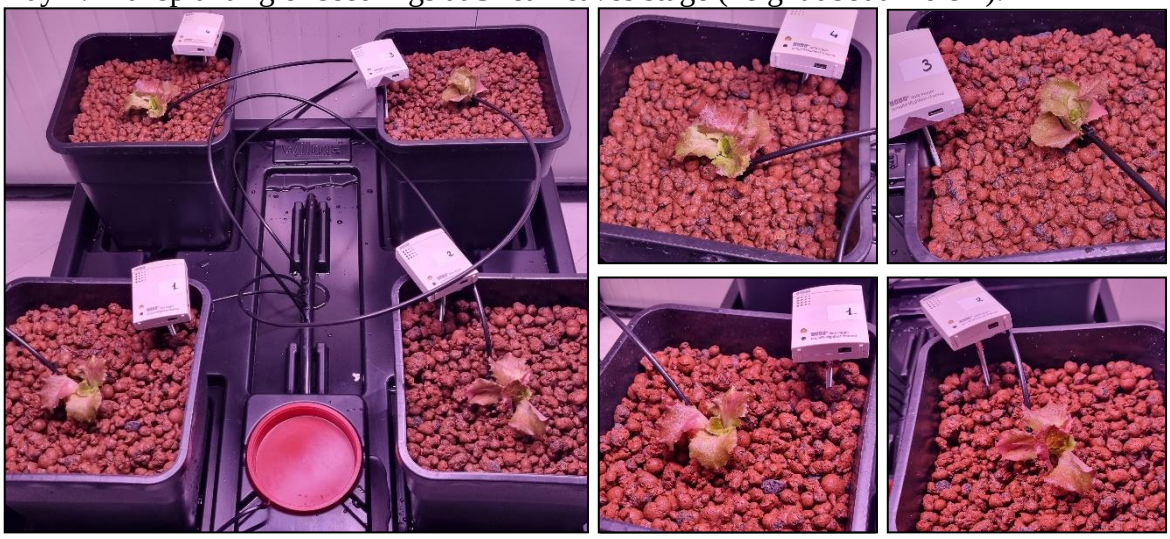
During the growth stages the following agronomic parameters were manually collected: root length, height of the head part, diameter, number of leaves, and final plant weight.

3.2.4 Results and Discussion

As for the previous microclimatic parameters characterization of the confined agricultural environment (see 4.1), the collected data were elaborated considering measured parameters under constant conditions. Thus, it is possible to assert that the standard deviation can be used to account for the uncertainty of the measured parameters and that the temporal mean of all the collected data by each probe represents a precise estimation of the parameters.

Below are reported the different stages of lettuce growth in the mini hydroponic system set-up over a period of 30 days.

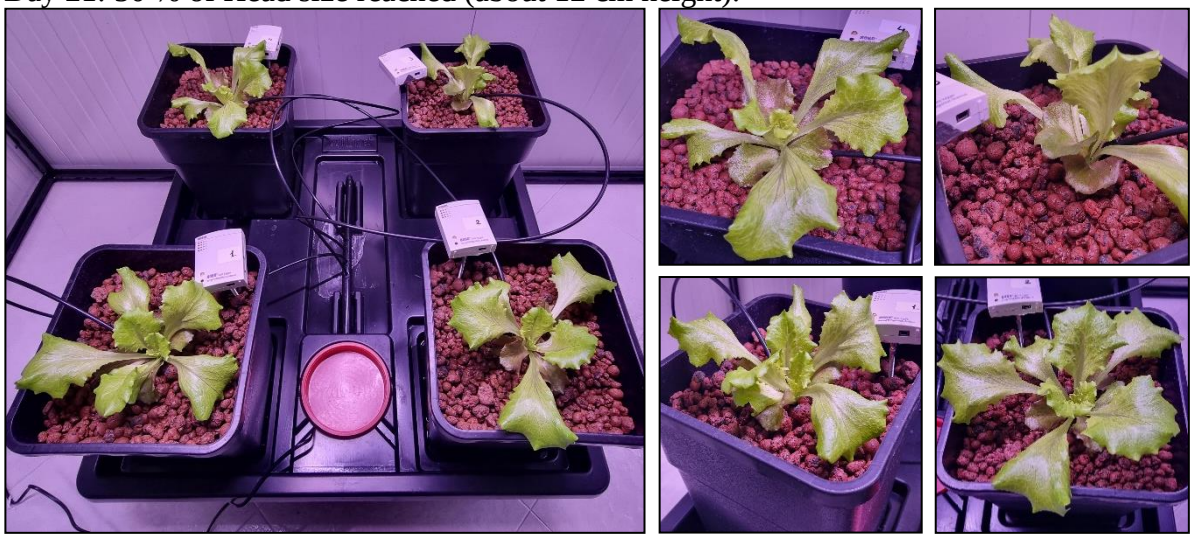
Day 1: Transplanting of seedlings at 3 real leaves stage (height about 4-6 cm).



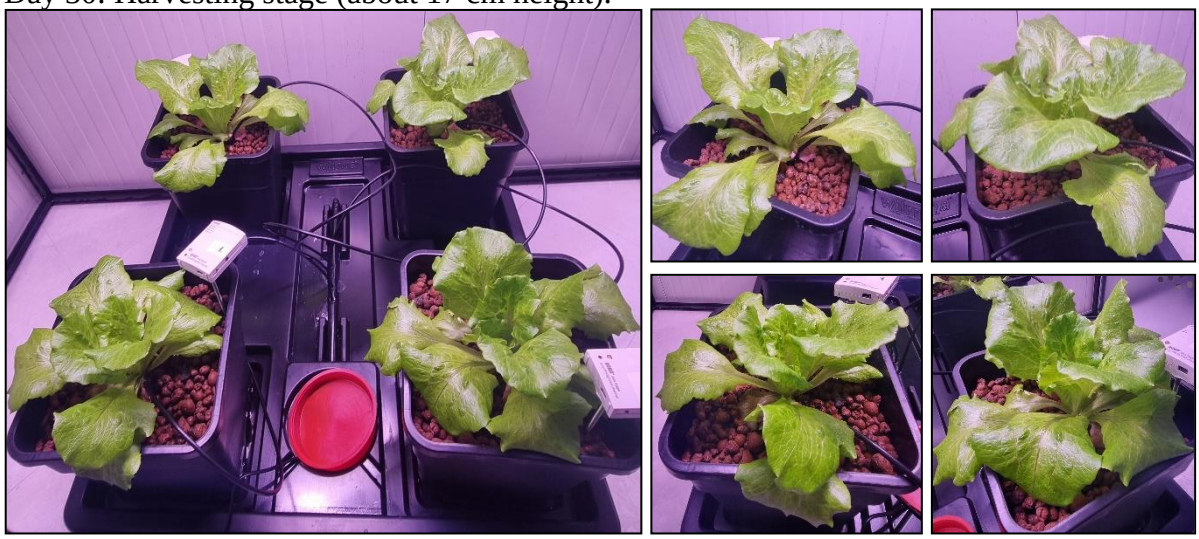
Day 7: 4-5 true leaves plant (height approximately 9 cm).

Day 14: head begins to form (about 13 cm height).

Day 21: 50 % of Head size reached (about 12 cm height).



Day 30: Harvesting stage (about 17 cm height).

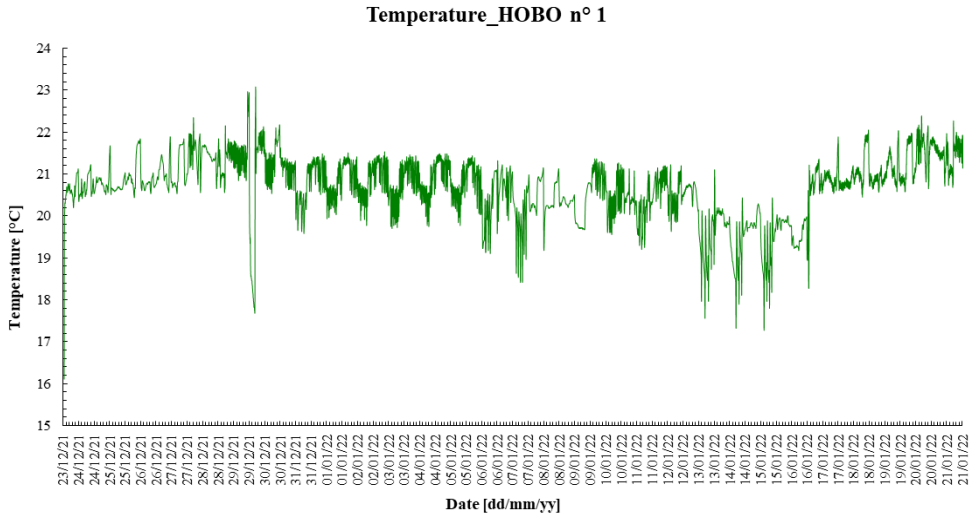


The following Table 13 shows the parameters acquired from HOBO for the entire growth phase.

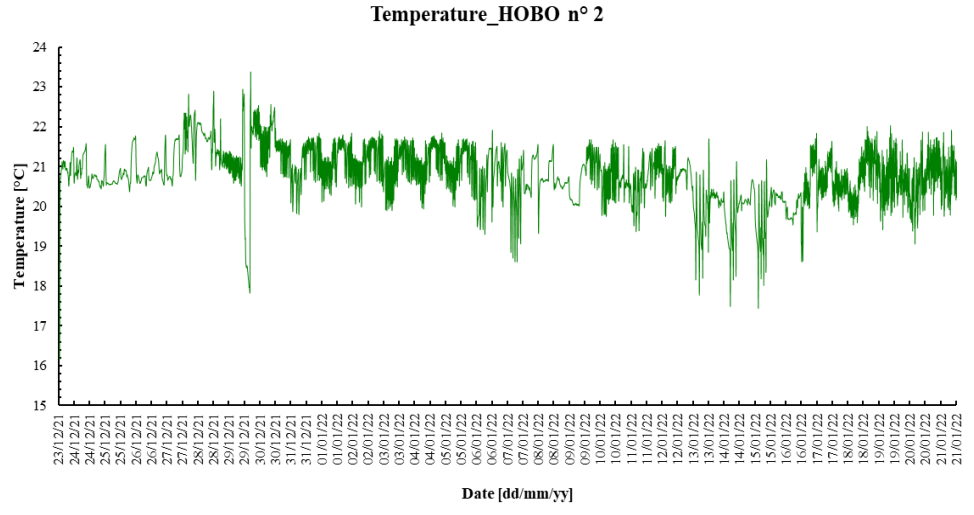
***Tab. 13**: Average values and standard deviation of measured parameters.*

		HOBO n° 1	HOBO n° 2	HOBO n° 3	HOBO n° 4
Temperature (T_{IN}) [°C]	$\bar{x}$	20.69	20.88	20.79	20.50
	σ	0.73	0.69	0.78	0.73
	$\bar{x}$		20.71		
	σ		0.36		
Relative Humidity (U_{IN}) [%]	$\bar{x}$	68.10	67.47	66.02	67.87
	σ	2.20	1.93	2.31	2.30
	$\bar{x}$		67.36		
	σ		1.09		

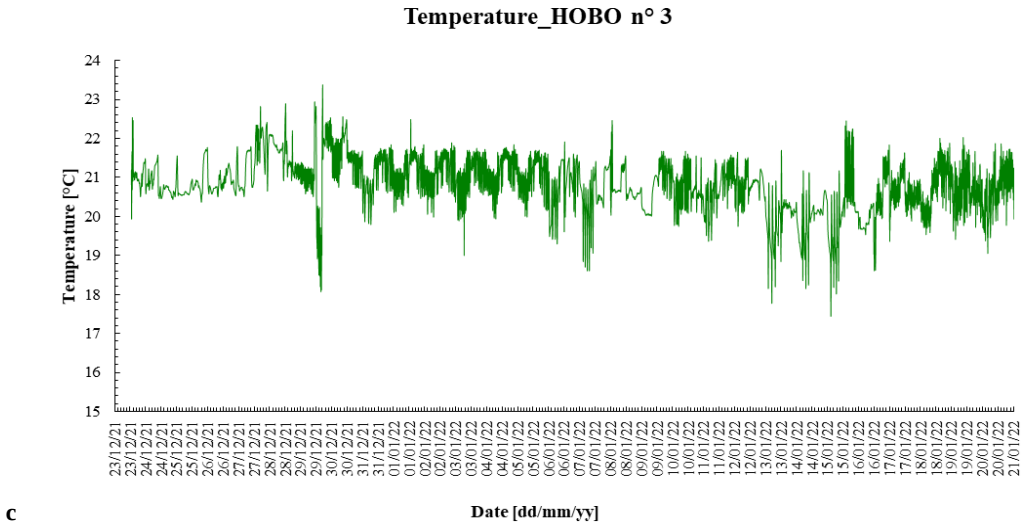
The average of the temperature T_{IN} was of 20.71 °C with a standard deviation of 0.36 °C. In general, temperature trends were homogeneous in each measured point as shown in Figures 30a, b, c, d.



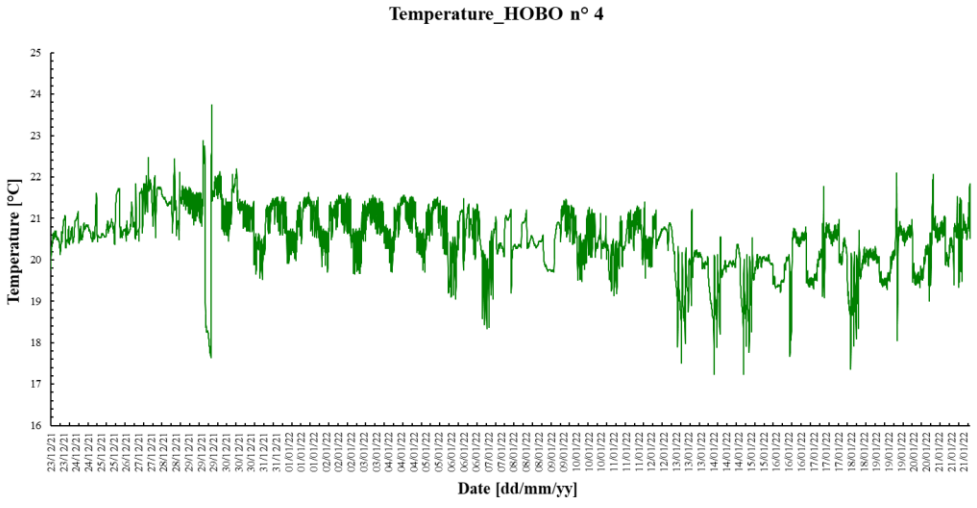
a



b



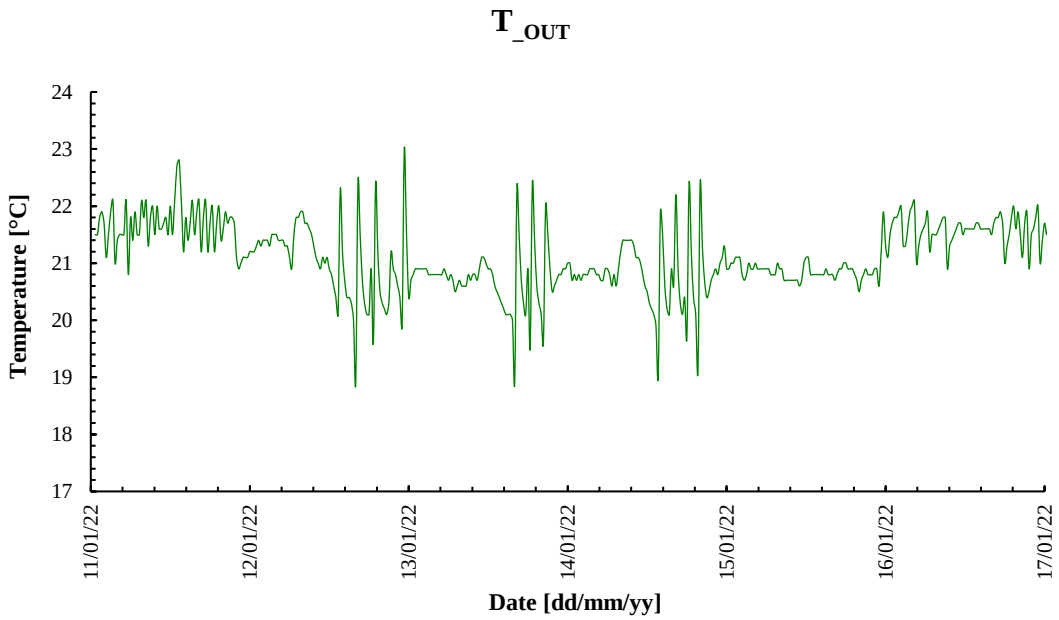
c



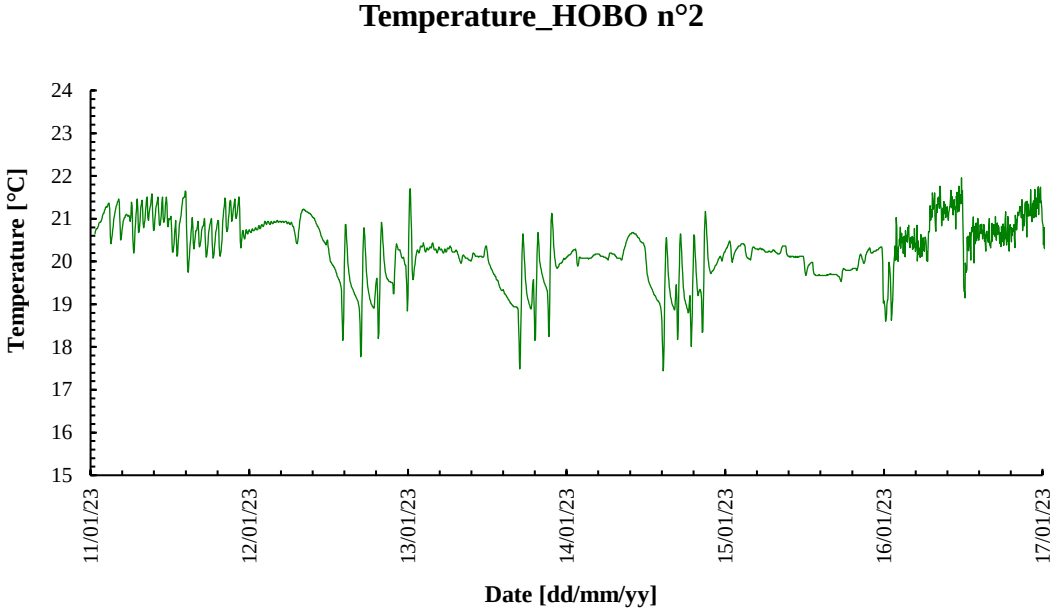
d

Fig. 30*: Temperature trends of four HOBO sensors in* ***a****),* ***b****),* ***c****),* ***d****).*

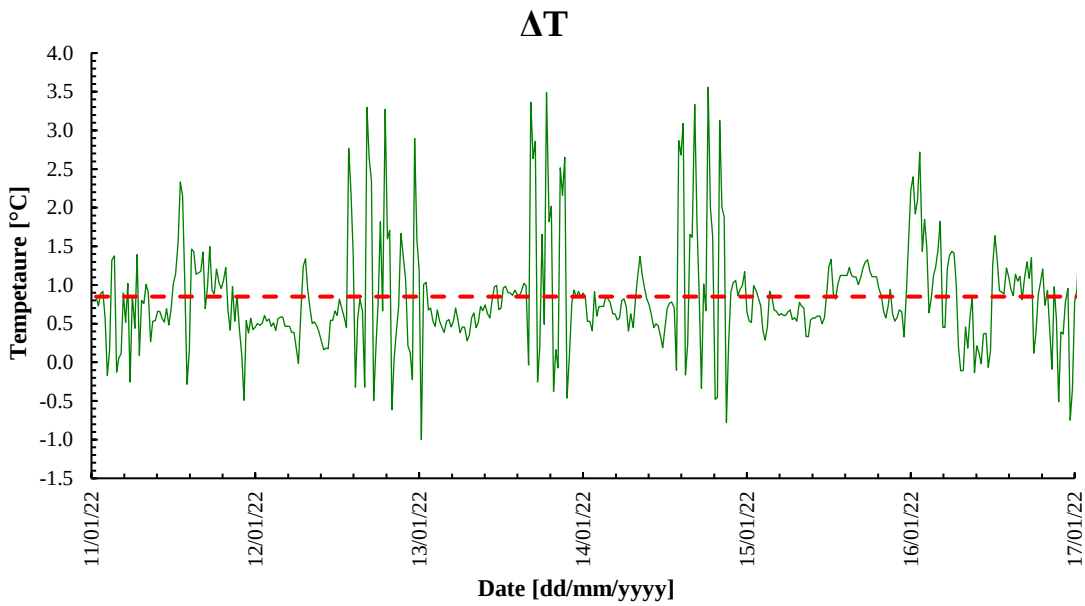
From the graphs few points reached a value of 17.5 °C due to door openings for the worker management operations. Nevertheless, this drop in temperature not affected the lettuce plant growth and confirmed that the experimental environment, if properly closed, is well insulated. The inlet air temperature near the holes $T_{\_OUT}$ recorded an overall average of 21.12 °C, with a standard deviation of 0.58 °C. There was a temperature loss of 1.94 % between the inside and outside of the box, but with a similar behaviour for each measurement. To further understand, the following Figure 31a, b illustrates a selected short time period from $T_{\_OUT}$ of 1 week in comparison with the same of a chosen $T_{\_IN}$, where this similarity in trend can be seen. To better highlight this trend, the temperature gradient ($\Delta T = T_{out} - T_{HOBO\ n°2}$) is shown in Figure 31c. It has an average value of 0.85 °C over the whole week and oscillates in the range between -1 °C and 3.56 °C. Thus, this confirms that the small scale environment showed a good insulation.


T_OUT
Temperature [°C]
24
23
22
21
20
19
18
17
11/01/22
12/01/22
13/01/22
14/01/22
15/01/22
16/01/22
17/01/22
Date [dd/mm/yy]

a


Temperature_HOBO n°2
Temperature [°C]
24
23
22
21
20
19
18
17
16
15
11/01/23
12/01/23
13/01/23
14/01/23
15/01/23
16/01/23
17/01/23
Date [dd/mm/yy]

b



c

***Fig. 31**: Comparison between the temperature at air inlet holes (**a**) and inside the cultivation box (**b**);temperature gradient between the outside and the inside of the box (**c**).*

Regarding the relative humidity, the $U_{\_OUT}$ had an average of 65.68 % with a standard deviation of 2.59 %, while the $U_{\_IN}$ nearby the plants had an average of 67.36 % with a standard deviation of 1.09 %. The $U_{\_IN}$ value was achieved for many factors: due to the isolation of the scaled growth box; due to the recirculation of the nutrient solution by the drip system; especially due to the fact that plants grew healthy and actively, thus transpiring more water. The result was a rapid increase in the water vapor content and humidity. In fact, it was recorded an increase in value of 2.6 %.

As for the temperature also $U_{\_IN}$ was uniform for each pot and Figure 32 represents an example case of the relative humidity trend (HOBO of plant number 4).

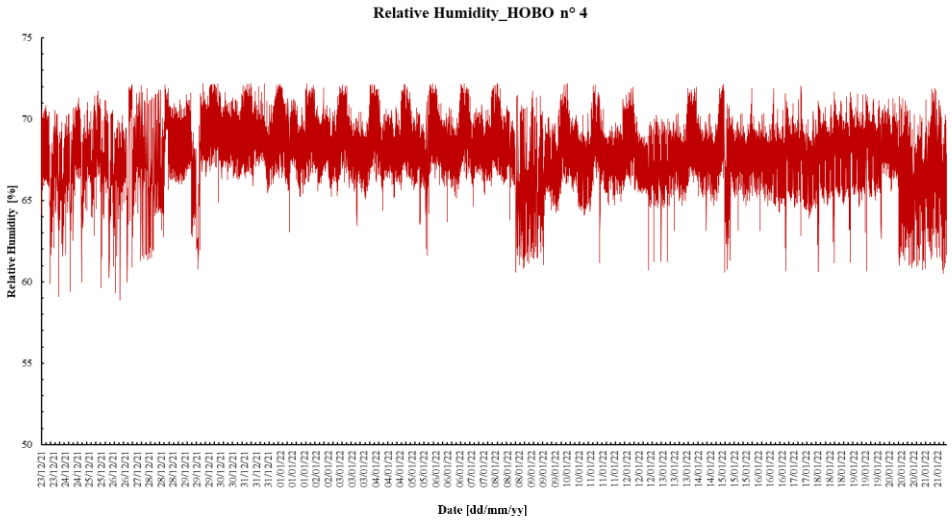


***Fig. 32**: Relative humidity trend of crop number 4.*

From the graph is possible to see how relative humidity is related to the temperature: at lower temperature corresponded higher relative humidity. In fact, air temperature decreases with no change in water vapor content, the maximum water-holding capacity of the air drops, resulting in a higher relative humidity. In addition, as for the temperature trend, also for relative humidity there were lower values compared to the average, reaching a value of about 60 %.

For each parameter observed the trends were consistent at night, which supports the suitability of the location where the hydroponic system was installed.

During the experimental test the inlet air flow rate at holes level was of 338.78 m^3/h, reducing to only 328.72 m^3/h in the growth box. CO_2 level was had a mean of 451.42 ppm and it did not required enrichment taking to account that the experimental test was conducted in the industrial area of Vinchiaturo (CB), Italy, well compatible with a big city where small hydroponics finds potential application. On the recovery grid, CO_2 value was of 553.49 ppm, indication that lettuce plants worked properly.

About the light intensity the mean value during the day-light cycle was of 2942.15 lux. From the lux average value was possible to get a DLI of 4.2 mol m^{-2} d^{-1} that also if low (minimum recommended DLI for indoor gardening systems for red-leaf lettuce plants is of 6.5 to 9.7 mol m^{-2} d^{-1}) it guaranteed a successful production. In next future it will be keep in mind to do test the cultivation at different under different day/night cycle schedule.

All these factors discussed above contributed to a successful plants growth, for which some agronomic parameters are reported in Table 14.

***Tab. 14**: Agronomic parameters measurements on lettuce seedlings.*

Phase	ID Plant	Roots Length (cm)	Head Length (cm)	Diameter (cm)	Leaves n°	Final Head Plant Weight (g)
Transplantation	1	4	6	5	3	23
	2	4	5.8	5.5	3	25
	3	4	4.7	6	3	24
	4	4	5.5	4.9	3	24.3
WEEK 1	1	/	8	7	6	/
	2	/	9	8	6	/
	3	/	9.5	7.2	7	/
	4	/	9.1	7.3	7	/
WEEK 2	1	/	13	15	7	/
	2	/	12.5	13	9	/
	3	/	11.6	12	8	/
	4	/	12	15.3	8	/
WEEK 3	1	/	15.5	21	11	/
	2	/	16.2	20	10	/
	3	/	15.4	21.6	11	/
	4	/	14.8	20.4	10	/
Harvesting	1	9.8	15.5	24	10.5	180
	2	9.5	16.2	29	15	190
	3	10	15.4	28	15.3	159
	4	10	14.8	25	10	168

From this table it is possible to note at first glance the root length, that are shorten compared to lettuce grew in a traditional hydroponic greenhouse (see Table in 3.1.2 Results and Discussion). Mean value of 12.7 cm is not an issue because there are many factors that contribute to its development: in this case roots are continuously irrigated with nutrient solution in an inert growing medium, thus they do not need to deepen towards the solution tank.

Moreover there was a consumption of about 10 litres of solution; the amount of nutrient solution used depends not only on the size of the grow area, but also on aspects as plant transpiration and evaporation rate. After the transplantation phase until the last cultivation week, plants were delicate and roots length was not collected to avoid damages. Consequently, also the final weight of the head part was postponed at the end, with a final head plant weight of each plant of about 180 g. This final weight varies according to

variety and is acceptable, especially if compared with that obtained in the hydroponic greenhouse.

3.2.5 Additional future applications

The experience of doctoral research and study abroad allowed to investigate about new features that could find application in the concept of mini hydroponic confined system. In particular, two developments might be interesting for implementing the system tested in this Ph.D. thesis.

The first application regarded the study of the application of LoRa radio communication technique to small controlled cultivation environments. It consisted of several phases: a first phase regarded the research, elaboration and acquisition of theoretical information about access technologies used for wireless data transmission and LoRa technology. During the second phase a market research of the components to be assembled was done, with a knowledge integration directly in field's workshops in Germany. The third phase consisted of the coding and testing phase of LoRa technology.

To better understand the work carried out, a short introduction of the context and of the concept of Lora is reported below.

Internet of Things regards a physical device network connected to the internet that are able to communicate each other. In addition, several wireless technologies can be used to connect these devices to internet: short-range wireless communications, Cellular communications and LPWAN (Figure 33). The latter stands for Low Power Wide Area Network and includes technologies like Narrowband, Sigfox and LoRa, that differ from each other for the relation between bandwidth and distance.

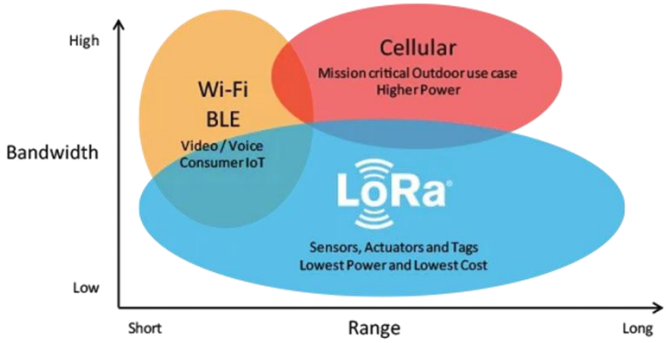


***Fig. 33**: Expected wireless data transmission ranges of technologies in relation to their bandwidth.*

Source: https://www.thethingsnetwork.org

LoRa is a wireless modulation technique derived from Chirp Spread Spectrum (CSS) technology, that is a long-range radio-frequency technology for wireless communication and consists of a chirp, a linear variation in frequency, that spans the entire band assigned to the channel. The following two tables (Tables 15a, b) give an overview of the main characteristics of some access technologies used for wireless data transmission.

***Tab. 15**: **a**) Frequency plans by Country of LoRa®; **b**) Main characteristics of some wireless data transmission technologies.*

Region	Frequency (MHz)
Asia	433
Africa (parts), **Europe**, India, Russia	**863-870**
US	902-928
Australia	433
Canada	863-870
China	902-928

a

Technology	Wireless Communication	Range	Tx Power
Bluetooth	Short range	10 m	2.5 mW
Wifi	Short range	50 m	80 mW
2G/3G/4G/5G	Cellular	5 km	5000 mW
LoRa®	LPWAN	2-5 km (urban area) 5-15 km (rural area) > 15 km (Direct line of sight)	20 mW

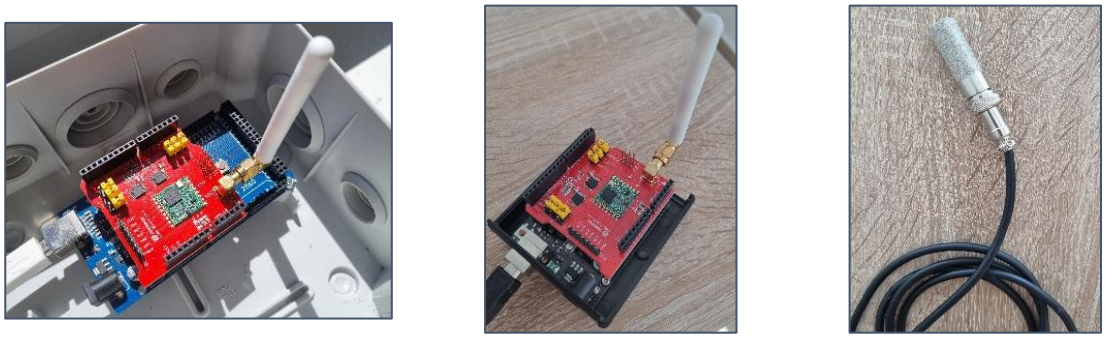
b

LoRa modulated transmission is robust against disturbances and the reception is over long distances. In fact, it sends small data packets with a data rates of 0.3 kbps-5.5 kbps at long distance from 2000 m to 15000 m on the base of the line of sight; the transmission power is of about 20 mW. LoRa uses the license of free sub-gigahertz bands: for example, 915 MHz, 868 MHz, and 433 MHz that are part of Industrial, Scientific and Medical (ISM) bands.

The application of LoRa technology was carried out at the Leibniz Institute of Agricultural Engineering and Bio-economy and concerned the Peer-To-Peer (P2P) LoRa Communication. In this type of communication each node communicates directly with the others without the mediation of a server.

For the testing phase the Peer-To-Peer (P2P) LoRa Communication between Lora Shield Sender and Lora Shield Receiver, among Lora Shield Sender, Lora Shield Repeater (more than one) and Lora Shield Receiver were obtained.

To carry this research out two LoRa module were assembled with the following components in Fig. 34a, b: Arduino UNO, MEGA, LoRa shields, and antennas. A Temperature-Humidity probe FS200-SHT20 (Fig.1c) was set and connected to test the system.



a **b** **c**

***Fig. 34**:LoRa sensors: **a**. Dragino LoRa Shield - 868MHz v1.4 + Arduino MEGA+ Antenna; **b**. Dragino LoRa Shield - 868MHz v1.4 + Arduino UNO+ Antenna; **c**. FS200-SHT20 temperature-humidity probe.*

Arduino.cc open-source electronic prototyping platform was used for the coding phase, setting the free bandwidth at 868 MHz, common for European Country and also specific for Germany. The codes were elaborated based on the specific cases and consulting several libraries on Github.com and on thethingsnetwork.org.

Below are shown in sequence Arduino's code written and tested for the Temperature-Humidity probe and for Lora Shield Sender, Repeater and Receiver.

Arduino Code for Temperature-Humidity probe:

```
/*
// Begin of Michela Orsino Temperature-Humidity probe Code
 Program name:  SHT20 Sensor probe
 Last update:   June.01.2022
 Author:       Michela Orsino, May.26.2022
 Product use SHT20 T-H Sensor probe
 Task 1:       Operation of the sensor probe
 // End of Michela Orsino Temperature-Humidity probe Code

 * Hardware Connections:
 * -VCC = 3.3V
 * -GND = GND
 * -SDA = A4 (use inline 330 ohm resistor if your board is 5V)
 * -SCL = A5 (use inline 330 ohm resistor if your board is 5V)
 */
#include <Wire.h>
#include "DFRobot_SHT20.h"
DFRobot_SHT20    sht20;
void setup()
{
   Serial.begin(9600);
   Serial.println("SHT20 Example!");
   sht20.initSHT20();                         // Init SHT20 Sensor
   delay(100);
```

```
    sht20.checkSHT20();                    // Check SHT20 Sensor
}
void loop()
{
    float humd = sht20.readHumidity();          // Read Humidity
    float temp = sht20.readTemperature();       // Read Temperature
    Serial.print("Time:");
    Serial.print(millis());
    Serial.print(" Temperature:");
    Serial.print(temp, 1);
    Serial.print("C");
    Serial.print(" Humidity:");
    Serial.print(humd, 1);
    Serial.print("%");
    Serial.println();
    delay(1000);
}
```

Arduino Code for Dragino Lora Shield Sender:

```
/*
// Begin of Michela Orsino LoRa Sender Code
 Program name:  Sender Device
 Last update:   July.01.2022
 Author:        Michela Orsino, June.10.2022
 Product use:   Dragino Lora node
 Processor:     Arduino Uno,Dragino Lora shield v1.4
 Task 1:        Peer to Peer communication with Dragino Lora shield Receiver
 // End of Michela Orsino LoRa Sender Code
*/
//------------------------------------------Begin Library-----------------------------------
#include <SPI.h>
#include <RH_RF95.h>
#include <DFRobot_SHT20.h>
//------------------------------------------End Library-------------------------------------
DFRobot_SHT20    sht20;
//-------------------------------------Begin LoRa Initialization-------------------------
// Singleton instance of the radio driver
RH_RF95 rf95;
//-------------------------------------------End LoRa Initialization---------------------
//-------------------------------------------Begin Setup---------------------------------
void setup()
{
  Serial.begin(115200);
  while (!Serial) ; // Wait for serial port to be available
  if (!rf95.init())
    Serial.println("init failed");
    sht20.initSHT20();                    // Init SHT20 Sensor
    delay(100);
    sht20.checkSHT20();                   // Check SHT20 Sensor
}
```

```
//----------------------------------------End Setup----------------------------------------
//----------------------------------------Begin of Loop----------------------------------
void loop()
{
  int sensor_value = analogRead(A0);
  Serial.println("Sensor n.1 Value : "+(String)sensor_value);
  uint8_t buf[RH_RF95_MAX_MESSAGE_LEN];
  uint8_t len = sizeof(buf);

  float humd = sht20.readHumidity();                  // Read Humidity
  float temp = sht20.readTemperature();               // Read Temperature
  String tempe = "Temp="+String(temp);
  String humid = " Humd="+String(humd);
  //String message = "I am No.1: " + tempe+humid;
  String message = "#1: " + tempe+humid;
  //Send a message to rf95_server
  uint8_t datam[30];
  message.toCharArray(datam,30);
  rf95.send(datam, sizeof(datam));
  delay(2000);
}
//----------------------------------------End of Loop----------------------------------------
```

Arduino Code for Dragino Lora Shield Repeater:

```
/*
// Begin of Michela Orsino LoRa Repeater Code
 Program name:  Repeater: Receiver&Sender Device
 Last update:   June.23.2022
 Author:        Michela Orsino, June.22.2022
 Product use:   Dragino Lora node
 Processor:     Arduino Uno,Dragino Lora shield v1.4
 Task 1:        Repeat message from Dragino LoRa shield Sender toward Dragino LoRa shield Repeater
// End of Michela Orsino LoRa Repeater Code
*/
//----------------------------------------------------Begin Library-----------------------------------------------------------
#include <SPI.h>
#include <RH_RF95.h>
//----------------------------------------------------End Library-------------------------------------------------------------
//----------------------------------------------------Begin LoRa Initialization-----------------------------------------------
//-----------------------------------Singleton instance of the radio driver---------------------------------------------------
//----------------------------------------------------End LoRa Initialization-------------------------------------------------
int led = 13;
//----------------------------------------------------Begin Setup-------------------------------------------------------------
void setup()
{
  pinMode(led, OUTPUT);
  Serial.begin(9600);
  while (!Serial) ; // Wait for serial port to be available
  if (!rf95.init())
  {
```

```
    Serial.println("init failed");
  }
  }
//------------------------------------------------------------End Setup-----------------------------------------------------------
//------------------------------------------------------------Begin of Loop-------------------------------------------------------
void loop()
{
   //---------------------------------Check if LoRa message is Available-----------------------------------------------------
  if (rf95.available())
  {
    // Should be a message for us now
    uint8_t buf[RH_RF95_MAX_MESSAGE_LEN];
    uint8_t len = sizeof(buf);
//---------------------------------------Begin of Check if LoRa message was received-------------------------------
//----if the receiver receives something, the following code run and the received data are saved in buf----------
    if (rf95.recv(buf, &len))
    {
      digitalWrite(led, HIGH);
          Serial.print("I am #3. Received from ");
      Serial.println((char*)buf);

      // add personal message to received data
      const int start = 4;
      char a[30];
      int i;
      for( i=0; i<sizeof(a);i++ ) {
         a[i] = 0x00;
      }
          char b[len-start];
      int k = 0;       // Initialize k
      // the for loop copies in the vector b the elements of buf starting from 14 to the end.
      // In this way, the first part of buf, "I am node #1: " is not resended.
      for (int i = start; i <= sizeof(buf); i++) {
        b[k] = buf[i]; // copy the element i of the string "buf" to "b"
        k = k+1; // increase l
      }
//-----------define a constant char called first with the message "I am #2 repeater. Message: "----------
      const char *first = "#3: ";
//---------------------------copy the string "first" in "a"-------------------------------------------------
      strcpy(a,first);
//---------------------------------attach the string "b" to "a"-----------------------------------------
      strcat(a,b);
//---------------------------------attach the string "buf" to "a"-----------------------------------------
      //strcat(a,buf);
      uint8_t len2 = sizeof(a);
//-------------------------Repeat the message-------------------------------------------------------------
      delay(100);
      rf95.send(a, len2);
//---------------------------------------Repeat the message-----------------------------------------------
      digitalWrite(led, LOW);
```

```
    }
    else
    {
      Serial.println("recv failed");
    }
  }
}
//------------------------------------------End of Loop-----------------------------------------------------
```

Arduino Code for Dragino Lora Shield Receiver:

```
/*
// Begin of Michela Orsino LoRa Receiver Code
 Program name:  Receiver&Sender Device
 Last update:   June.23.2022
 Author:        Michela Orsino, June.22.2022
 Product use:   Dragino Lora node
 Processor:     Arduino Uno,Dragino Lora shield v1.4
 Task 1:        Peer to Peer communication with Dragino Lora shield Sender: Receive message from the Sender
 // End of Michela Orsino LoRa Receiver Code
*/
//------------------------------------------Begin Library---------------------------------------------
#include <SPI.h>
#include <RH_RF95.h>
//------------------------------------------End Library-----------------------------------------------
//-------------------------------------Begin LoRa Initialization-----------------------------------
// Singleton instance of the radio driver
RH_RF95 rf95;
//-------------------------------------End LoRa Initialization-------------------------------------
//------------------------------------------Begin Setup-----------------------------------------------
int led = 13;
void setup()
{
   pinMode(led, OUTPUT);
  Serial.begin(9600);
  while (!Serial) ; // Wait for serial port to be available
  if (!rf95.init())
  {
    Serial.println("init failed");
  }
}
//------------------------------------------End Setup-------------------------------------------------
//------------------------------------------Begin of Loop---------------------------------------------
void loop()
{
  if (rf95.available())
  {
    // Should be a message for us now
    uint8_t buf[RH_RF95_MAX_MESSAGE_LEN];
    uint8_t len = sizeof(buf);
    if (rf95.recv(buf, &len))
```

```
    {
      digitalWrite(led, HIGH);
      Serial.print("I am #2. Received from ");
      Serial.println((char*)buf);
      Serial.print("RSSI: ");
      Serial.println(rf95.lastRssi(), DEC);

      // Send a reply
      //uint8_t data[] = "And hello back from you";
      //rf95.send(data, sizeof(data));
      //rf95.waitPacketSent();
       digitalWrite(led, LOW);
    }
    else
    {
      Serial.println("recv failed");
    }
  }
}
//-------------------------------------------End of Loop----------------------------------------------
```

From the work carried out data were successfully transmitted and repeated from a device to another.

The P2P communication was very good and was also strictly dependent on power supply of devices.

This code running permitted to transmit Temperature-Humidity data from a Dragino Lora Shield to a Repeater that, in turns, retransmitted them to another Dragino Lora Shield Receiver. Figure 35 shows the working system; in particular, data transmission and reception are displayed from the serial ports interfaces COM5 and 9.

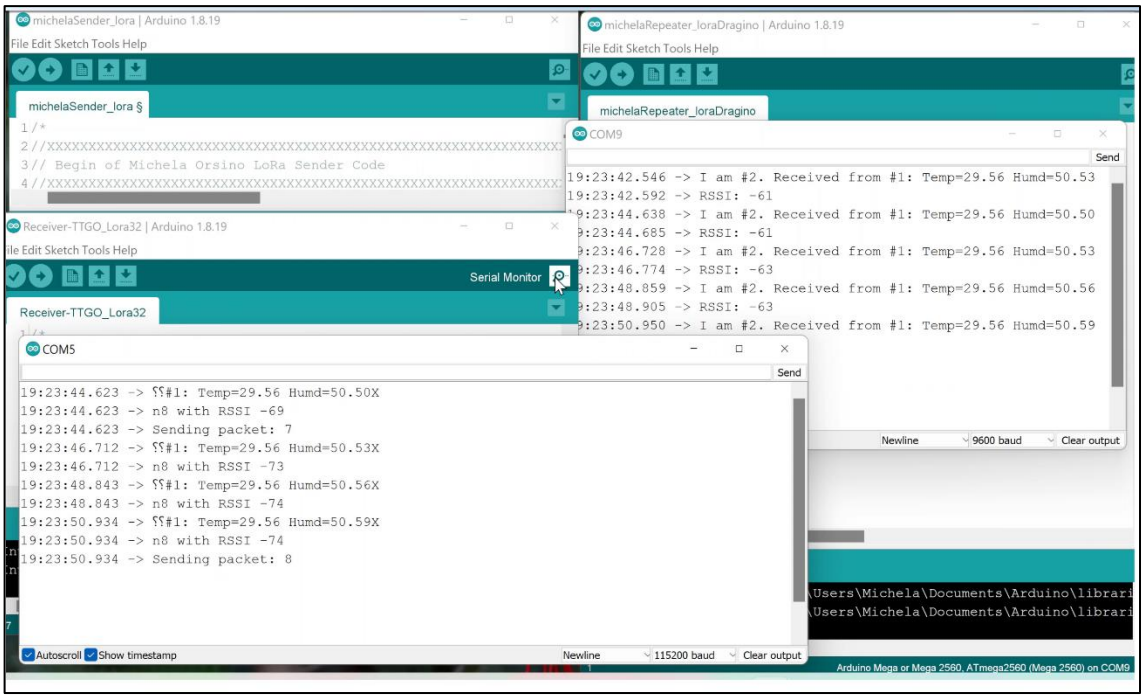


***Fig. 35**: Data transmission from the Sender, connected to the sensor probe, to the Receiver (COM9), via the Repeater (COM5).*

The sensor probe is interchangeable and, if properly coded, it can work well with this system. The devices were tested in a urban range, at around 2 km of Direct Line of Sight.

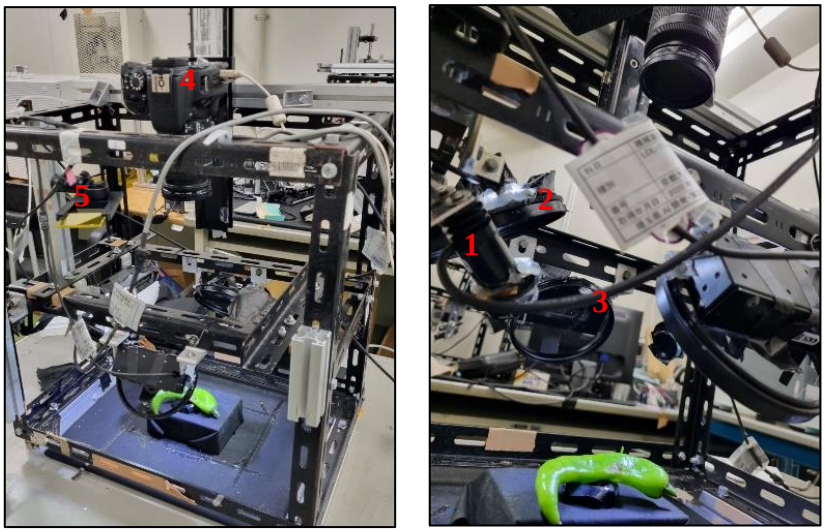
Due to time limitation, as next future perspectives the aim of the work is to test the system at >15 km of Direct Line of Sight, fixing sensors in the small hydroponic system and for remotely monitoring the cultivation.

The second possible future application concerned the study and the application of spectroscopy techniques as fluorescence imaging for food evaluation. During the brief period at the Bio-sensing engineering Laboratory of Kyoto University in addition of learning about fluorescence imaging for food quality evaluation, there was the opportunity to participate in a first part of an experimental test on Manganji sweet pepper for early “blossom and rot” (BER) detection.

Manganji Togarashi is a valuable Japanese variety of green peppers, but it is particularly affected by BER (caused by calcium deficiency in the early growth stage and which turns the color of some parts of the pepper from green to brown up to yellow) during summer, resulting in its devaluation on the market. Based on the idea of early detection of this disease, I learnt a lot about the practical and general application of fluorescence imaging system as an effective method to measure fluorescence compounds in real-time and both destructively and non-destructively.

The experiments were based on the excitation-emission matrix (EEM) measurements, that is a destructive method using a JASCO FP-8300 spectrofluorometer. The spectrofluorometer measured, in the wavelength range between 250 nm and 750 nm at 5 nm intervals, the light intensity of the fluorescence emitted by the sample when illuminated by an excitation light field whose wavelength varies between 235 nm and 735 nm at 5 nm intervals. This represents an effective method for estimating the fluorescence compounds of food products. The samples were produced by a front face method, i.e. by cutting the sample from the Manganji pepper. Indeed, the front-face method provides a direct measurement in which absorption, reflection and scattering phaenomena are drastically reduced, greatly improving the results. This measurement gives a three-dimensional data matrix containing the values of excitation wavelength, fluorescence intensity, and emitted fluorescence wavelength. From these values, it is possible to determine the presence of the disease and its characteristics.

Based on the EEM results, fluorescence and colour images of the samples were captured using a purpose-built system (Figures 35a, b) composed by a Canon EOS DSLR kiss x7 camera, and three types of LEDs: 365 nm UV LED + Y49 blue light filter, 420 nm UV LED + G455 filter to filter blue light and produce a weak halation, and White LEDs.



a **b**

Fig. 36*: Set up of image acquisition system:* ***a****) 1) 420 nm UV LED, 2) White LED, 3) 365 nm UV LED, 4) camera Canon EOS DSLR, 5) polarizing filters.*

In general, the 365 nm LEDs are used as an excitation source for detecting surface fluorescence of green peppers, because they provide a wide excitation wavelength band (from 345 nm to 395 nm) that is suitable for these purposes.

From the figures below (Fig. 36a, b, c) it is possible to observe how BER color, defined by the emission wavelength, changes under different LEDs, i.e. under different excitation wavelengths.

a **b** **c**

Fig. 37*: Image of Manganji peppers illuminated by:* ***a)*** *White LEDs, where the BER area is darker,* ***b)*** *365 nm LEDs+Y49, where the BER area is yellowish,* ***c)*** *420 nm LED+ G435, where BER area is brighter with halation because of 3D shape change.*

MATLAB software was used for data processing and, in particular, to extract: R (red), G (green), B (blue) pixels values, R, G, B ratio, and HSV (Hue Saturation Brightness), L* (the perceptual lightness), a*and b*(the chromaticity coordinates that stand for the four unique colours of human vision: red, green, blue and yellow). All images were labelled manually as BER area and Health area as shown in Figure 37.

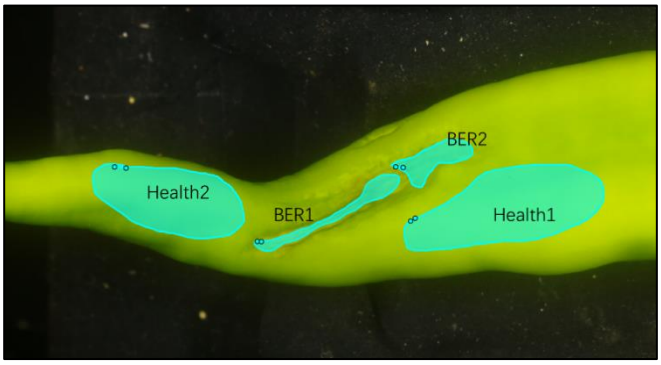


Fig. 38*: Color image segmentation in region of interest: BER area: BER1; Possible BER area: BER2; Health area (1 cm close to BER area): Health1; Health area (1 cm close to BER area): Health2.*

The region of interest (ROI) was manually delimited using color images and, then, applied on the fluorescence images by overlaying color images on the corresponding fluorescence images. Initially, the original values were in the RGB color space, then converted to L*, a*, b* and HSV and the mean values of individual color channels were calculated for the analysis.

The activity at Kyoto University was functional to doctoral project because the fluorescence imaging techniques can find application in controlled agricultural environment. In fact, it was considered the realization of a scaled down version of this

apparatus (see previous Figure 35) to insert in the cultivation chamber, as a non-destructive solution for a real time monitoring of the vegetable production. Thanks to this system it could be possible to optimize the production preventing possible disease for the crops, due to biotic and abiotic factors.

4 Conclusions

In the present Ph.D. thesis, a fully closed small-scale soilless cultivation system for lettuce with artificial light was designed, realized and studied. Beside its developments, the work consisted of other experimentations that were functional to the main project.

In order to compare the microclimatic monitoring tests performed in the mini controlled environment agriculture, a microclimatic data acquisition was carried out in a large standard hydroponic greenhouse. Acquired data were elaborated to evaluate the trend of the microclimatic parameters inside the hydroponic greenhouse and to identify the best optimization asset. In particular, two microclimatic monitoring studies were conducted for this work in a hydroponic greenhouse. Tests were conducted on lettuce Salanova, differentiating two areas: Z areas for the cultivation tank and C areas for the central aisle. The first monitoring test, during the springtime, highlighted issues with high relative humidity levels, particularly on days whenever it rained; in fact, from one rainy day to the next sunny day there was an increase of even 20 % of relative humidity. In the second monitoring test, conducted in the summer, the indoor humidity conditions improved due to both increased temperatures and the installation of a mechanical fan, with a 12 % and 30 % reduction in relative humidity on the cultivation side and 30 % lower relative humidity in the central aisle, respectively. This reduced the onset of diseases and also had a positive impact on phytopathology. Even though the maximum temperature in the greenhouse was around 34 °C, the average daily temperature in the side cultivation zones and in the central aisle dropped by around 15 % in comparison to the first test. This demonstrated the fan's efficacy. Based on the results of these two microclimatic tests, it is evident that the structure should be improved because it is not properly closed and insulated (day/night temperature range) and that a mechanical ventilation system that automatically regulates temperature and humidity is required to reduce the fluctuations identified by the set of sensors used in this work.

In addition, the best way to improve technology as mini hydroponic system is through the characterization of microclimatic characteristics in confined agriculture environments. To further improve its development, it was essential to enhance the microclimatic control. Thus, before the realization of the small scale hydroponic system, a preliminary study to evaluate the distribution of the microclimatic parameters in the confined agricultural environment conditioned was necessary to be carried out.

The air conditioning system was handled by ensuring the microclimatic parameters of a 40 m^3 CEA for successive hydroponic lettuce plants cultivation. A microclimatic control unit, which simulates the plant, was used to achieve this goal.

Tests were carried out fixing the set-points, suitable for lettuce cultivation, for temperature and relative humidity at 21 ± 1 °C and at 70 ± 5 %, respectively, and performing the measures of temperature, humidity, globe thermometer temperature, and air speed at different fan speeds (30 %, 50 %, 80 %) for air conditioning supply. Their were under control by means of a handling unit, which provided the treated air through a perforated duct, and a humidifier.

The main results demonstrate that, in all tests, the controlled environment was characterized by good spatial homogeneity of parameter values, ensuring the same conditions for all the plants regardless of their location. However, the optimal conditions were obtained at 80 % of fan speed, corresponding to 10 air exchanges per hour. This is due to increased turbulence, which leads to better water vapor mixing and more even temperature distribution. The air speed near the crop seemed to be unaffected by speed, remaining constant at 0.1 m/s in all cases, demonstrating that the air handling device was properly sized. Additionally, it has been demonstrated that using perforated duct is a highly effective way to uniformly distribute microclimatic parameters inside the confined agriculture environment. Finally, this system's reliable environment microclimatic management makes it possible to guarantee a quicker and more effective lettuce cultivation.

After the preliminary investigation of microclimatic parameters distribution, the aim of the work was the realization and study of a hydroponic cultivation system in scale, completely confined. Four lettuce plants were monitored during one production cycle of 30 days thanks to a network of sensors, which can work both offline and cloud-based, to monitor the cultivation system real time. A constant daytime indoor temperature of 21 °C, with a dead band of ± 2 °C and a relative humidity set-point of 65 %, with a dead band of ± 5 %, were fixed as set-points and were regulated by the sensor probe at air inlet holes.

In general, temperature and relative humidity trends were homogeneous in each measured point.

However, it was reached a temperature value of 17.5 °C in few points due to door environment openings for the worker management operations. However, this not affected the lettuce plant growth and confirmed that the experimental environment was well

insulated. Also in this case is evident how relative humidity is related to the temperature: also for relative humidity there were lower values compared to the average, reaching a value of about 60 %.

Finally, this system's reliable environment microclimatic management makes it possible to guarantee a quicker and more effective lettuce cultivation.

Research is therefore promising and can be used in the development of industrial products.

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