Tampere University of Applied Sciences

Responses of Sweet Basil to Varied Daily Light Integrals in Germination, Yield, and Morphology within A Plant Factory

Kieran Denham

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ABSTRACT

Tampereen ammattikorkeakoulu
Tampere University of Applied Sciences
Energy and Environmental Engineering

DENHAM, KIERAN:
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Vertical farming is an unconventional agricultural practice that brings fresh food production into the urban environment. Controlled greenhouse style environments fitted with artificial lighting, with the added feature of vertically stacked layers, provide the ideal architecture for fast and efficient plant growth. Stable climate and light conditions provide reliable output for the farmer, the drawback is often the high electrical demand and advanced technology that is required to make the system efficiently viable.

The objective of this work is to study the function and theory behind the artificial irradiance and physical environmental factors that affect sweet basil growth (*Ocimum Basilicum L.*) inside a Closed Plant Production System / Plant Factory. This provides insight into responsive variation of the crop that can then be utilized to inform better operation of such an existing Plant Factory.

The methodology follows the growth of 23 pot sown sweet basil plants by manual measurement of whole plant leaf areas, fresh weights, dry weights, heights, and photosynthetic active radiation levels at 33-days growth period as exposed to varied levels of irradiance (PPFD). The collected data was regressed against functional plant characteristics such as leaf area index, fraction light interception, leaf area ratio, and daily light integrals.

The results in this report showed that insufficient light levels caused stunted growth in selected samples. With the combination of comparatively high light levels in other samples, high variation in growth rates across all samples could be seen. Linear relationships between leaf area index and fresh weight, leaf area index and dry weight, and leaf area index and height were clear through manual measurements. Lower range light treatment and higher range light treatment during the germination period did not satisfy plant growth requirements.

Key words: vertical farming, PFAL, sweet basil, LAI, DLI

CONTENTS

ABBREVIATIONS AND TERMS

CPPS – Closed Plant Production System
DLI – Daily Light Integral
FLI – Fraction Light Intercepted
LAR – Leaf Area Ratio
LT-A – Light Treatment A
LT-B – Light Treatment B
LT-C – Light Treatment C
LT-M – Light Treatment Module
LED – Light Emitting Diode
PAR – Photosynthetic Active Radiation
PFAL – Plant Factory with Artificial Lighting
PPFD – Photosynthetic Photon Flux Density
Module – The production part of the PFAL in which the plants receive photosynthetic light and are transported horizontally along the production layer.

1 INTRODUCTION

The United Nations Food and Agricultural Organization (2018) reveals that since the 1960's the worlds available arable land per person is slowly decreasing, and by 2050 will become one-third the amount available since 1970. This is the affect of many factors including climate change, bio-fuel energy demands, urbanization, desertification, water scarcity, and, perhaps most clearly, because of population growth (FAO 2009; Fedoroff 2015).

Vertical farming is the consequence of the demand by the urban metropolis for both very high supply of fresh produce and a very low physical presence, saving increasingly precious arable land area. Urban agriculture has been a long tradition in people's lives and is considered integral to resilient and sustainable 'city region food systems'. (FAO 2014.) In Latin America, for example, many countries such as Cuba with 40% of households and Guatemala with 20% of households practice Urban and Peri – Urban agriculture.

A vertical farm or PFAL (Plant Factory with Artificial Lighting) refers to a food production facility with a thermally insulated and near air-tight warehouse-like structure (Kozai 2013). Inside, vertically stacked crop layers are fitted with electric photosynthetic lamps that, along with other necessities, promote fast and efficient plant growth. Lamps, such as Light Emitting Diodes (LED's) that can produce light energy for plant uptake, are increasingly being used in PFAL's owing to their low surface temperature, compact size, high light use efficiency, and broad light spectra (Kozai, Niu, & Takagaki 2016; Ashdown 2015; Ahmad et al. 2018). The primary function of these LEDs is to satisfy the requirements of leaf optical properties that include different chlorophyll pigments to capture light energy and initiate the photosynthesis process (Avgoustaki, Li, & Xydis 2020).

Plants both affect and are sensitive to light quality and quantity (Aphalo & Ballare 1995). Crop quality and yield in an open field or greenhouse are subject to seasonal differences in sun radiation and outside temperature. (Heuvelink, Ooster, & Stanghellini 2019, 279.) Even in advanced greenhouses incident light is mostly unregulated and so these differences still exist to an extent, therefore an open market is available for high quantity and quality of product year-round in PFAL, and the like, structures. Prediction of growth and development of the crop is important for efficiency and quality, ultimately benefitting a more marketable and desirable product. Within a PFAL the available level of control allows the operator to dictate the amount of energy directed towards particular elements of stress within the system.

This report is a response to the desire for a better understanding of the growth capabilities of sweet basil (*Ocimum Basilicum L.*) crop cycles within an existing PFAL. In particular, the response of the sweet basil crop to varying light intensities during germination and throughout the plant's growth till harvest. The functioning PFAL used was a novel multi-layer semi-automated herb and micro-green year-round Closed Plant Production System (CPPS) with an effective ground surface area capacity of $70m^2$ (7m x 2.5m x 4 layers). The PFAL in use, namely The Little Garden ®, was built in 2018/19 in Metropolia University of Applied Sciences Myyrmäki Campus, Finland. Ideally, the client would like to establish a total growth period (including germination) of 30 days or less with a production output that has low variation between plants and across batches and is of high quality. This would mean, for the business, the farming of sweet basil in this environment could be an economical and reliable crop rotation. The experiment also aims to show, quantitatively, variation in yield as a response to varying Daily Light Integrals primarily across the germination period but also throughout the main growth period in the vertical farming context.

In this report, the concepts that affect the quality and quantity of the crop will be introduced. The theory of Photosynthetic Active Radiation (PAR), light spectrum, and Daily Light Integral (DLI) is presented and is necessarily deployed in the development and growth analysis. To understand and evaluate the effect of varying light intensities on basil growth there are several factors that must be monitored and controlled. Both air temperature and humidity of the PFAL and crop canopy, and carbon dioxide concentration were measured at several light intensities. Plant Leaf Area's (cm^2), Fresh Weights (g), and Dry Weights (g) were manually measured at the end of a 33-day growth period.

2 THEORY

2.1. Vertical farming

PFAL's and CPPS's are drivers of innovative design in agricultural practice in the face of an expected 34% increase in the world's population by 2050, a steady decline in crop yields, and increased urbanization up to 70% by 2050 (FAO 2009). Technological feats and efficiencies have "made viable a commercial approach for the large-scale production of a variety of crops in close proximity, or even within, urban centres" (Despommier 2013), and so 'building upwards' provides the chance to bring together optimal conditions for growth and crop density closer to city centres. Within CPPS's there is also no agricultural 'run-off' therefore large water savings can be made (70-80%) (Despommier 2013). PFAL's run entirely on artificial lighting and therefore electrical energy consumption is generally the most energy intensive part of the process. Light efficiency is therefore especially important as only a fraction of the light energy emitted by modern LED's are absorbed and converted into chemical energy fixated by plants (Figure 1). The value at each step indicates the percentage of electrical energy consumed.

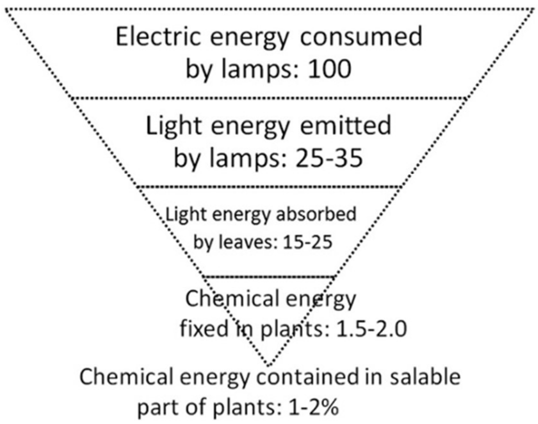


FIGURE 1. The need for light efficiency: Pyramid of energy efficiency under artificial lighting. Adapted from Kozai, Niu, & Takagaki (2016, 25) with permission by the Publisher*.

* This article was published in Plant Factory: An Indoor Vertical Farming System for Efficient Quality Food Production, Vol 1, Kozai T, Niu G, & Takagaki M, Figure 2.21, pge 25, Copyright Academic Press (2021)

Quality of the light source can be attributed to the light spectrum which for plant photosynthesis and morphology is within the visible wavelength spectrum 380-780nm (often 400-700nm). (Kozai et al. 2016.) Quantity of the light source is the light intensity which can be measured in radiometric units of absolute energy (W m^{-2}), photometric units of footcandle (lx), or quantum units of photons per area per second (µmol m^{-2} s^{-1}). Additionally, the total amount of light per day is described as Daily Light Integral (DLI) and is measured in mol m^{-2} d^{-1}. DLI is based on the light intensity or PAR and the photoperiod which takes account of the night schedule when the LED's are turned off. Photosynthetic efficiency is the amount of Dry Mass (DM) produced per unit of absorbed energy. (Natr & Lawlor 2005.) This absorbed energy is derived from how effectively the crop canopy is absorbing solar radiation. It is through the plant surface leaf area of the canopy, growing period and environmental conditions that promotes this effectiveness.

2.2. Physical environment conditions

As mentioned earlier, growth rate can be explained mainly by the ability of the crop to intercept and utilize the radiation at the canopy. This involves the leaf surface area of the plant and the DLI. The presence of light notwithstanding, the major factors that influence the growth rate is temperature, humidity, and CO_2 (Figure 2).

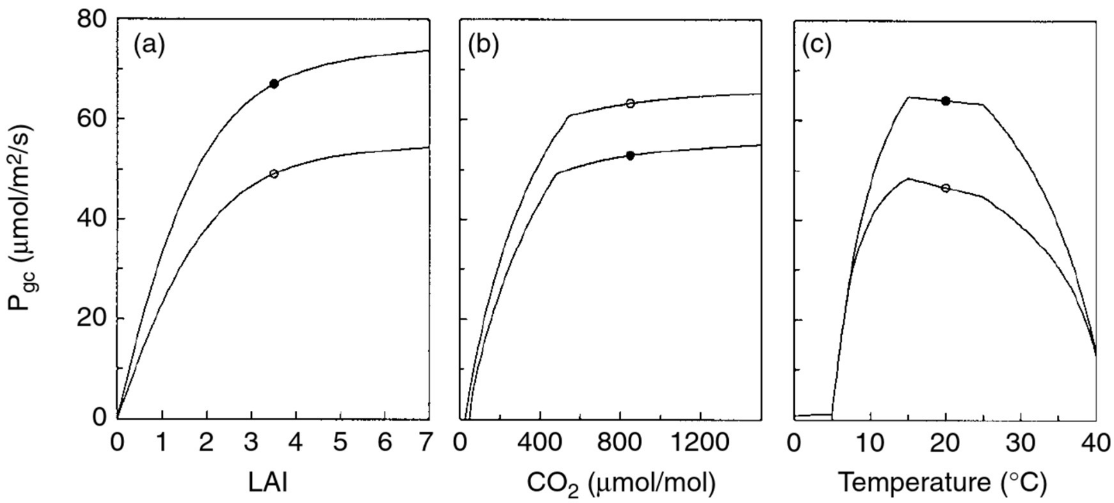


FIGURE 2. Simulated crop photosynthesis at 1500 μmol/m^2/s diffuse light as a function of (a) LAI: (at CO_2 concentration o = 340μmol/mol and ● = 1000μmol/mol), (b) CO_2 concentration (at o = 20^{o}C and ● = 30^{o}C), and (c) temperature (at CO_2 concentration o = 340μmol/mol and ● = 1000μmol/mol). Taken from Tomatoes (Heuvelink 2005, 89). Reproduced with permission of CAB International through PLSclear.

Temperature is one of the most influential factors on the development of plants and the operation of PFAL's (Heuvelink 1989; Kozai et al. 2016; Heuvelink et al. 2019). Figure 2 (b) shows the decrease in crop photosynthesis at higher and lower temperatures outside the optimum temperature ~25^{o}C (Kozai et al. 2016, 153).

Raising of CO_2 levels in greenhouse air stimulates crop growth considerably. (Heuvelink et al. 2019.) Elevated CO_2 causes higher production of assimilates, resulting in more branches, thicker leaves, and better fruit set. Temperature dependant, Heuvelink, Körner, & Niu (2009, 233) showed a ~50% increase in gross crop photosynthesis at higher CO_2 concentrations in greenhouses.

Humidity is the measure of the content of water vapour in the air. (Kozai et al. 2016.) Relative Humidity (RH%) is temperature-dependant and expresses the amount of water vapour the air can hold based on air temperature and pressure. Leaf transpiration rate is affected strongly by the surrounding air and its' moisture concentration. (Kozai et al. 2016, 156.) Leaf transpiration rate effects nutrient uptake as it is connected with mass flow and Ca uptake.

2.3. Available space

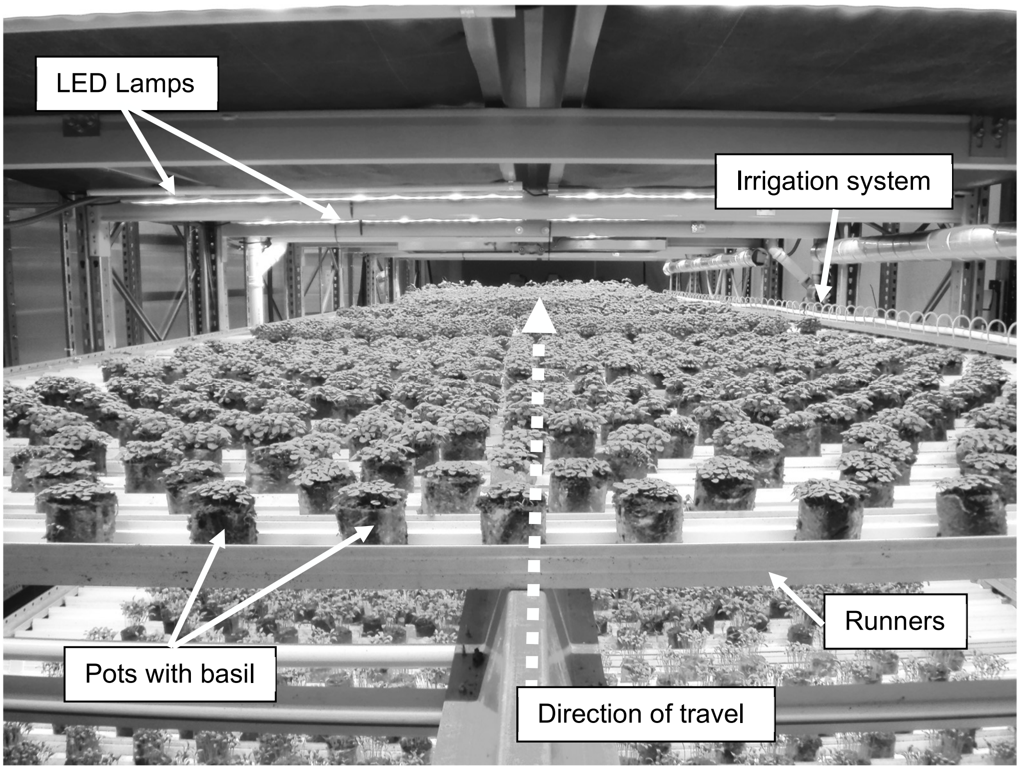
The continual growth and expansion of the basil canopy impacts on the adjacent plants. This is because of a combination of the design of the module, the arrangement of the plants within the runners and the encroachment of leaves laterally into other plants. A distance is set between batches and between plants within batches, therefore a maximum desirable LAI is required so to reduce the impact of inter-plant competition and provide even growth throughout the batch.

2.4. PFAL Structure

The Little Garden ® is a self-contained structure *within* a building that has also its' own air conditioning, lighting, etc. for office space. The PFAL structure houses all the necessary elements for normal operation. The PFAL is not airtight and therefore air and moisture exchanges are made between the indoor building conditioning and the PFAL.

2.4.1 Shell and layers

A UV-blocking polymer sheeting ‘shell’ encloses the PFAL growing layers. This shell is not built to be water or airtight, there are few outlets for electricals and gaps in the doors that air can freely move between the building and the inside of the PFAL. The growing layers or module layers (Picture 1) consist of a drip type irrigation system that feeds water into 2.5m long shallow gutters or ‘runners’ that sit laterally throughout the layer. The runners are hand sown with peat and seeds and then placed within the module where a pneumatic system ‘drags’ them longitudinally along the layer.



PICTURE 1. PFAL set-up with basil pots within runners set into the production layer.

2.4.2 Lighting

Parus (c) Water cooled LED grow lights (Parus n.d.) are set laterally to the module layer in sets of 2 x 10 (20 LED’s per layer). They are a Red-Blue-Standard light spectrum LED construct (Figure 3: bottom right) that can produce up to 735μmol/m^{2}s @ 0.5m with a light array angle of 130^0. Canopy photosynthetic capacity of basil was found to be saturated at ~500µmol m^{-2} s^{-1} at 25 ± 4°C by Beaman, Gladon, & Schrader (2009).

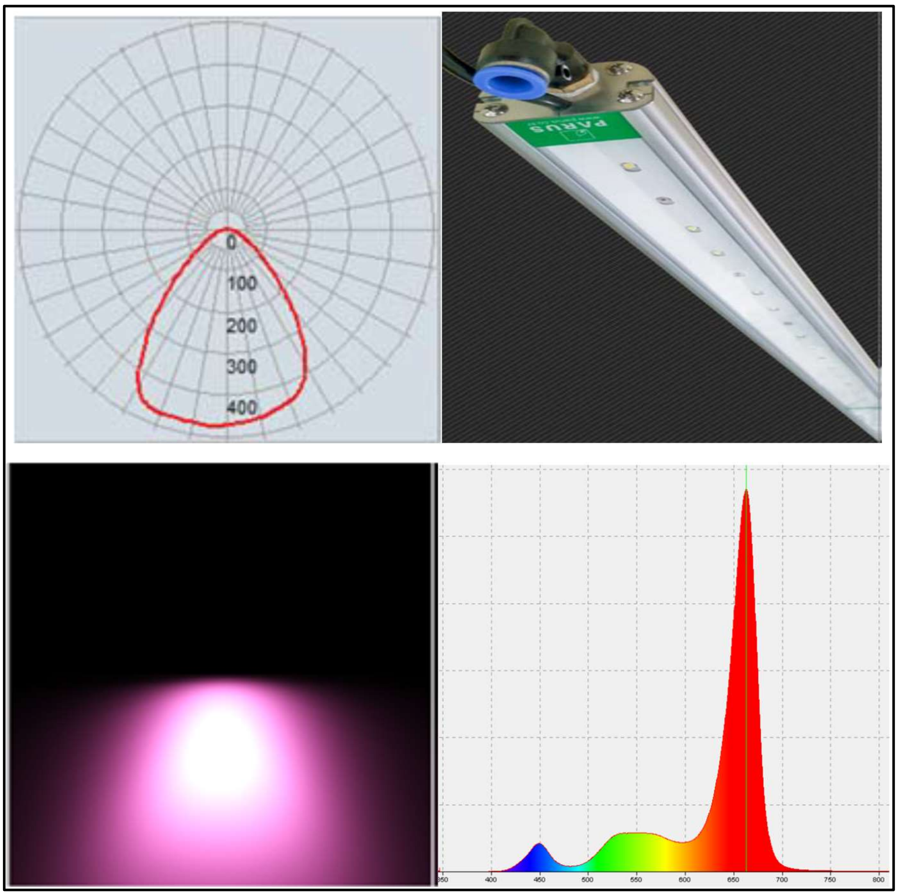


FIGURE 3. Parus (n.d.) water cooled LED grow lights. Top-left: Modelled light array. Top-right: water cooled LED lamp. Bottom-left: Image of light array. Bottom-right: Light spectrum of RBS LED.

The height of the basil canopy as well as the position within the module is continuously changing throughout the growth period i.e., the basil grows taller towards the lamps. Therefore, the distance between the LED's and the crop canopy is decreasing. The LED's were set at a continuous light intensity and photoperiod for the duration of the growth period so a gradual increase in the PAR is present at the canopy. Changes in LED light array angles are considered a key parameter in simulating radiative transfer and energy and mass balance of vegetative canopies (Wang, Li, & Su, 2007). This means that measurements at the soil or plant surface does not describe accurately the Photosynthetic Photon Flux Density (PPFD) at the canopy at any given time. Measurements at a set height e.g., at the soil level, however, are a good indication of the baseline PAR variance across the x-plane that will reveal ‘low light’ areas that may affect individual growth activity. Similarly, this unveils the extinction factor of the light toward the edges of the module. Simulated PAR arrays as experienced at the soil level is visualized in Figure 4.

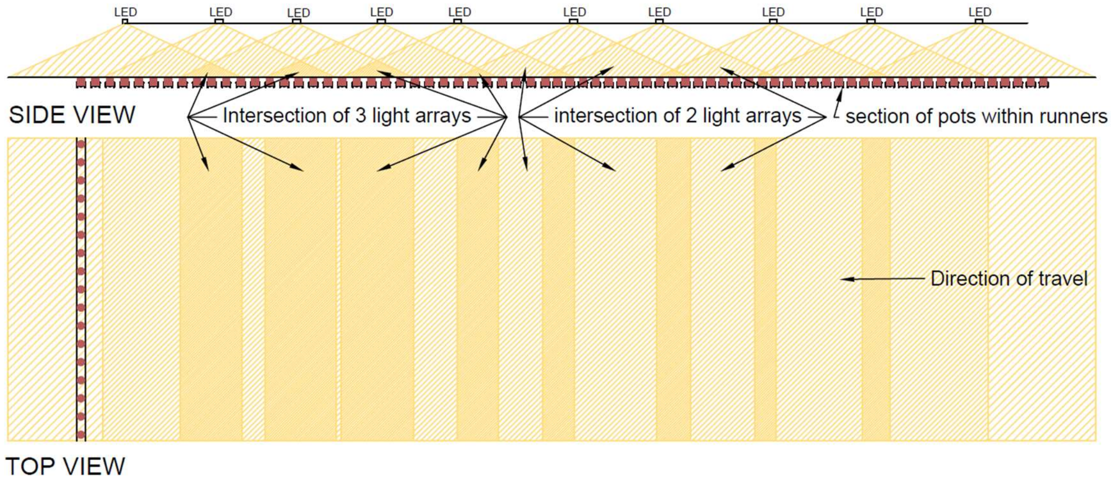


FIGURE 4. Simulated PAR array intersection across layer.

The least dense yellow hatched region in Figure 4 shows the basic presence of a LED’s PPFD, the denser cross hatching represents the crossover from the adjacent LED arrays.

2.4.3 Light spectrum:

There are minor variations in the photosynthetic effectiveness of photons of different wavelengths (Heuvelink et al. 2019). Experiments made by Dou, Niu, & Gu (2019) and Paradiso, et al. (2011) using green and purple basil cultivars and rose leaves respectively, showed increased lower-level plant canopy photosynthesis in the crop with the introduction of higher levels of green light in the PAR spectrum. Red and blue light, however, have the most important wavelengths for plant biomass accumulation by affecting plant photosynthesis and photomorphogenesis (McCree 1972 & Paradiso et al. 2011). Choong, He, Liu, & Qin (2015) reveal that Chinese cabbage (*B. Abloglabra*) grown under a Red-Blue combination LED increase leaf area and shoot FW by 36 – 121% and 34 – 119% respectively as compared with monochromatic blue light. It was also concluded that a red-blue ratio of 84:16 was the most effective for photosynthetic activity.

2.5. Development and Growth Analysis

The main factors that influence growth are light and CO_2 (Heuvelink et al. 2019, 27). The light energy (PAR) as experienced by the crop, is characterized by the DLI (the product of PPFD and photoperiod). This represents the total amount of light received by the crop in a 24-hour period. (Kozai et al. 2016) DLI can be simply calculated by reducing PAR by a daily integral.

$$DLI = PAR/1\ 000\ 000 * 60 * 60 * t$$

Where; *t* is the number of hours per day that the LED's are running constant PAR.

Leaf Area (LA) is the total on-sided surface area of the basil leaves' photosynthetic tissue using the method as described by Bazaz, Karimian, & Bannayan (2011) (see Methods), measured in cm^2. Leaf Area is an important driving variable for plant growth, as only light intercepted by green leaf area results in photosynthesis and growth (Heuvelink et al. 2019). Ground area is the surface area of the pot, which in this case was ~25cm^2. In young plants the growth is exponential in nature (Natr & Lawlor 2005, 506), in comparison with linear and slowing down of growth at the middle and end respectively during a plant's life. This is known as sigmoidal growth (Lee et al. 2003). Growth rate is therefore relative during this phase (Relative Growth Rate, RGR) and can be characterised by the Net Assimilation Rate (NAR) and the Leaf Area Ratio (LAR) described by the following:

$$RGR = NAR * LAR$$

Where;

$$NAR = 1/A * dW/dt$$

$$LAR = A/W$$

where A is the leaf area (cm^2), W is plant fresh weight (g), and t is time (days).

LAI is a dimensionless variable and was first defined as the total one-sided area of photosynthetic tissue (LA) per unit ground surface area (Watson 1947). It is 'a measure of the total photosynthetic system' (Monteith 1977) and is therefore considered a fundamental attribute of the canopy structure (Parker 2020).

$$LAI = LA/Ground\ area$$

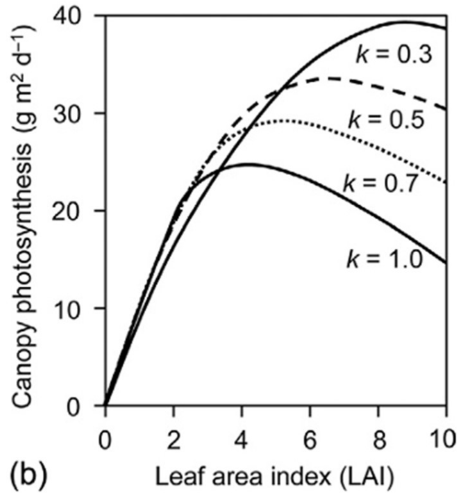


FIGURE 5. LAI as a function of canopy photosynthesis. Adapted from Kozai et al. (2016, 147) with permission by the Publisher*.

When extinction coefficient – k is approaching 1.0, beyond an LAI of 4, Figure 5 shows that canopy photosynthesis or crop growth per day can decrease since insufficiently irradiated lower leaves have higher loss of assimilates due to respiration than gains as a result of irradiation.

LAI and *k* follow Beer-Lamberts law of light extinction (Lai et al. 2012), that is;

$$I = I_o e^{-kLAI}$$

$$k = -\frac{1}{LAI}\left(ln\frac{I}{I_o}\right)$$

Where, *I* is the measured in-situ PAR inside the plant canopy, I_o is the measured in-situ PAR at the canopy level, *k* is the extinction coefficient, and *LAI* is the Leaf Area Index. Small values of k imply that less of the total LAI participates directly in light attenuation and absorption. (Parker 2020.) Related to the extinction coefficient is the Fraction Light Intercepted (FLI) represented as a fraction of the light intercepted by the canopy (Figure 6).

$$FLI = 1 - \frac{I}{I_0}$$

* This article was published in Plant Factory: An Indoor Vertical Farming System for Efficient Quality Food Production, Vol 1, Kozai T, Niu G, & Takagaki M, Figure 9.5 (b), pge 147, Copyright Academic Press (2021)

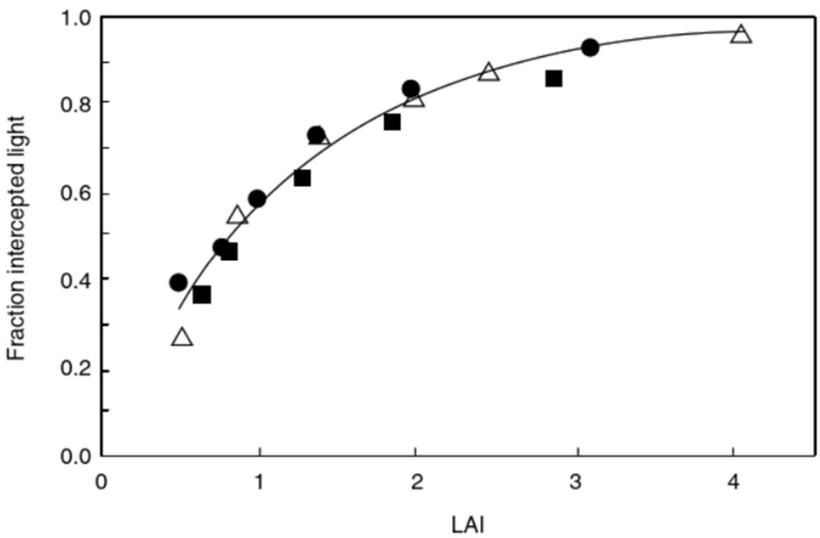


FIGURE 6. Light interception of young tomato plants arranged at different plant densities in order to vary leaf area index (LAI). Taken from Tomatoes (Heuvelink 2005, 88). Reproduced with permission of CAB International through PLSclear.

Figure 6 clearly shows that an inverse exponent relationship is present between LAI and FLI, wherein FLI reaches a limiting value at around LAI = 4. That is; as the plant canopy Leaf Area increases the amount of light penetrating to the lower levels of the crop is decreasing exponentially and has a maximum around 95%.

2.6. Marketability

In simple terms, there is a minimum and maximum height requirement that must be met by the basil if it is to be packed and sold to the marketplace. Pre-made marketplace packages dictate these limits. If the basil is too short, then the product is undesirable in-stores as it seems small in comparison to the packaging. If the basil is too tall, it will rise above the packaging and risk being torn during transport or while being stocked. A maximum of 30cm and a minimum of 22cm is required.

3 MATERIAL AND METHODS

The experiment was conducted within a walk-in LED vertical garden system (PFAL) run under the company name Little Garden Oy within Metropolia University of Applied Sciences Campus in Myyrmäki, Helsinki, FI, using Sweet Basil 'Ocimum Basilicum' from Enza Zaden (Netherlands).

3.1. Experimental setup and herb monitoring

Monitoring of herbs began 28.1.2021 until 2.3.2021 which took note of the development of 24 pot sown basil plants across varying light conditions. There were two phases in the monitoring of the plants. The first phase begins with the basil pots arranged within plastic racks as they arrive from the manufacturer. This is also how they are arranged during sowing and the germination stage. Phase 1 also follows three separate light treatment schemas as discussed further below. The second phase includes the main growth of the basil as they are moved from phase 1 locations and arranged within the vertical garden module layer. Both phases involve variations in DLI, water schedule and volume, temperature, carbon dioxide concentration, and humidity.

After the germination period (4 days) the pots were photographed and placed into the module growing layer that is dedicated to basil growth (phase 2). Each runner that holds the pots can hold 16 pots with a 15 cm gap from centre to centre between each pot (Picture 1). For practical reasons it was required to establish the maximum and minimum amount of growth that basil would undergo by the end of the growth period, this was validated by the arrangement of the pots within the runners. Further details are outlined below in section 3.2 Light treatments. After the 33-day growth period the plant heights were measured then cut at the base. FW, DW, and total LA of pot plants were then measured. A 'limiting' set of plants was then identified based on several factors including the height, quality and their corresponding plant characteristics: DLI, FW, LA, LAI, and climate details.

3.1.1 Climate

During both phases the air temperature, relative humidity, and CO2 were recorded approximately twice per week using the Elcometer 319 Dew Point handheld meter (Picture 2) which measures air temperature, RH% and surface temperature, and the Vaisala CO2 Probe GMP252 that measures CO2 in ppm. Continual monitoring equipment is also installed, it consists of a custom-made meter that has installed a Winsen MH Z19 Infrared CO2 sensor (±1ppm) and an Eiechip DHT21 Temperature and Humidity sensor (±0.1^{o}C, ±0.1%). Both measurements are made to make note of any major discrepancies and reliability of the data. Additionally, the air temperature and relative humidity within the basil canopy, once it was practical to do so*, was recorded (~twice per week).

Air temperature is controlled in a few ways; firstly via a heat pump at the ceiling of the PFAL, secondly via water cooling of the LED lighting system, thirdly a set of fans circulating the air through the basil layer from one end, and finally through heat exchange between the PFAL cell walls and the buildings indoor climate. The heat pump is an active control measure that is set to maintain 24 degrees Celsius within the whole module and the water cooling is latent and runs water through a small pump and heat exchanger that is pre-set to keep the lamps below a certain temperature for the sake of integrity and heat sink property. The fans together effectively introduce a flow of air and CO_2 into the basil canopy. The climate conditions present during the experiment are outlined in Table 1.

* after approximately 12 days growth period the basil canopy had developed sufficiently to place the meter within the stems.

PICTURE 2. Handheld Elcometer 319 Dew Point measurement examples. Left: Elcometer measuring air temperature and RH% within the plant canopy, Right: Elcometer measuring air temperature and RH% above the canopy within the module (Denham 2021)

Root Mean Squared Error (RMSE) is the standard deviation of the residuals in the regression analysis. RMSE was used to evaluate how well the manual climate measurements matched with the automatically monitored climate measurements.

$$RMSE = \sqrt{[\Sigma(Pi - Oi)^2/n]}$$

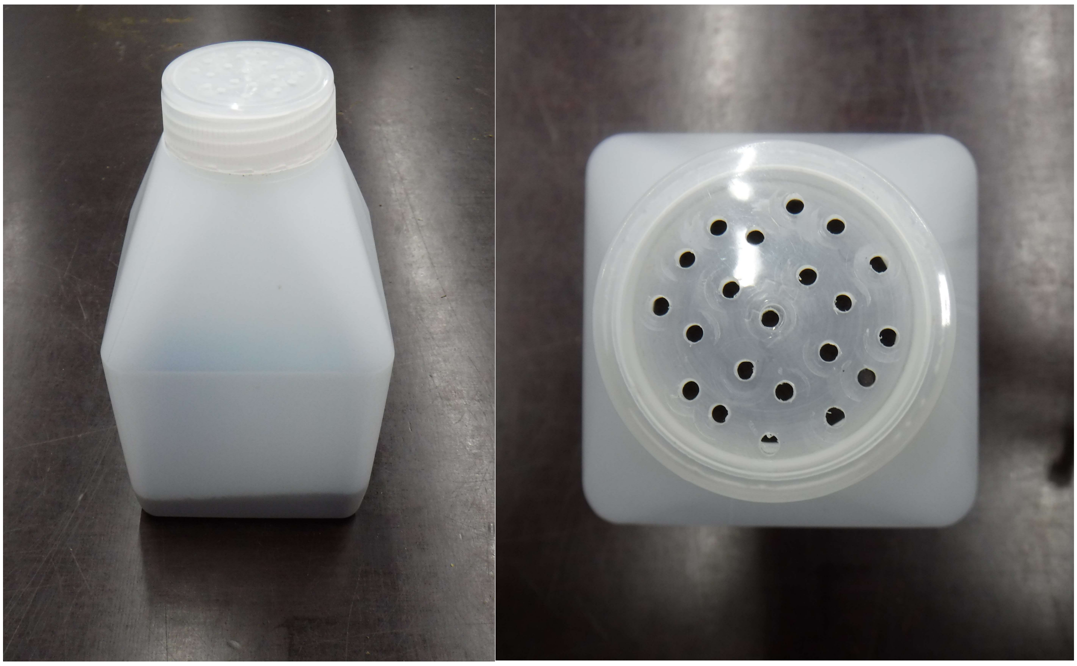
TABLE 1. Climate Conditions

Phase #	T_{air} (°C)	RH (%)	CO_2 (ppm)
1	23.9 ± 0.5	74.8 ± 1.7	512 ± 10
2	21.8 ± 1	76.8 ± 5.7	510 ± 14

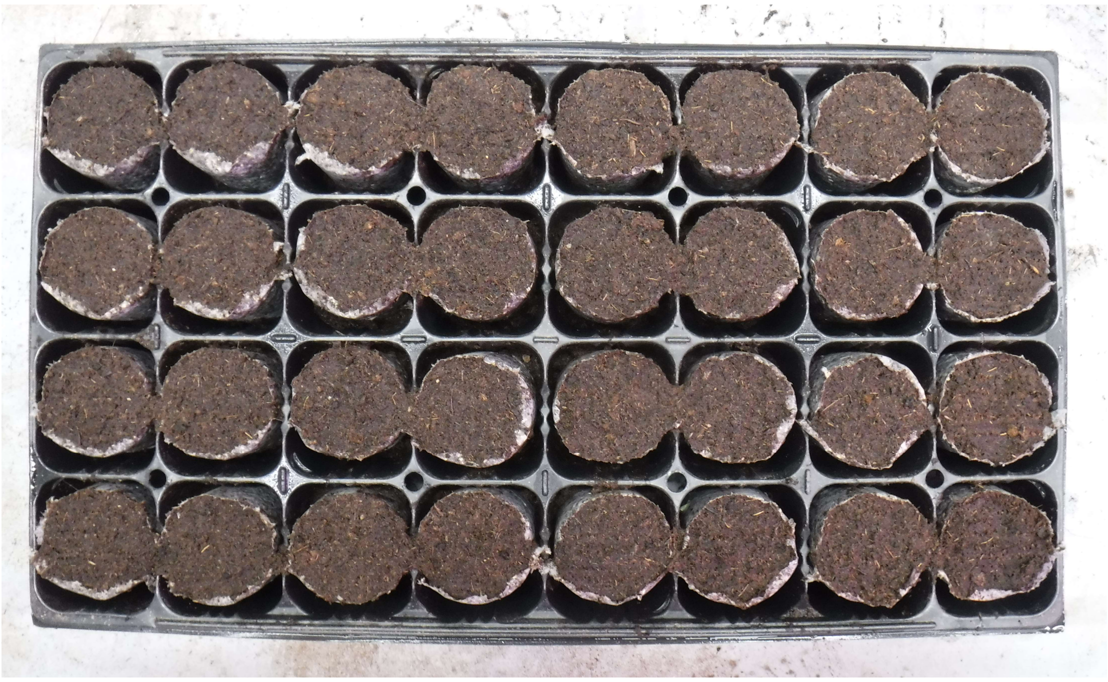
3.1.2 Sowing and Germination

Sowing and germination is carried out within the plastic racks for easy handling. The pots come from Robberts (c) soil processing operation in Southern Finland. For all cultivars, the method of sowing followed that which had been in operation at The Little Garden already. Basil seeds were held in a container with a colander type cap (Picture 3). The container/ dispenser is held over the plant pot and shaken until an approximate seed count is evenly dispersed over the surface of the pot (see Picture 4: top).

The desired rate of seed distribution was 50-70 seeds/ pot. This was estimated by measuring the seed dispenser weight before and after sowing and dividing by the number of sown pots. The average weight of 50 seeds was measured using a laboratory scale (n=5, 0.01g error margin).



PICTURE 3. Dispenser for basil seed sowing (Denham 2021)



PICTURE 4. Plant pots made from peat/ soil mixture approximately 175cm^3 inside plastic rack as they arrive from manufacturer (Denham 2021)

PICTURE 5. Left: germination has begun ~24h after sowing. Right: ~48h after sowing (Denham 2021)

3.2. Light Treatments

The two phases involve varying light intensity schema or Light Treatments (LT). The first phase exposes the basil to three different light treatments that reflect their location within the PFAL space. Phase 2 involves the arrangement of the germinated/ pre-grown basil pots within the PFAL module itself (Table 4) for the remaining of the basil growth before harvest.

3.2.1 Phase 1

At the germination stage until approximately 1 weeks' growth the combined height of the pots and basil are less than 10cm tall and do not require the physical space or the full light intensity as provided by the PFAL. Therefore, as a space saving measure, three locations A, B, and C situated outside the module layer are used during germination. The final location during phase 2 is within the module growing layer referred to as Light Treatment M (LT-M).

Each LT in the germination stage consists of 8 basil pots (8*3 = 24 in total), each sown and arranged as is described in the Sowing and Germination section above. A DeltaOHM HD 2302.0 LightMeter was used to measure the PPFD at different points across the trays holding the pots. The meter operates via a handheld computer attached to an illuminance photon reader (μmol m^{-2} s^{-1}) for wavelengths

400-700nm. The phase 1 LT PPFD ranges are shown in Table 2 and LT time schedule is shown in Table 3.

TABLE 2. Phase 1 light treatment PAR and DLI ranges.

Light treatment	PAR range (µmol/m2s)	DLI (mol/m2d)
LT-A	12.26-17.35	0.75-1.06
LT-B	30-46.2	1.84-2.83
LT-C	85-113.7	7.34-9.82

TABLE 3. Light treatment time schedule

Light treatment	Light period	Dark period	Total photoperiod per day	Days running
LT-A	07:00 – 24:00	00:01 - 06:59	17h	4
LT-B	07:00 – 24:00	00:01 - 06:59	17h	4
LT-C	00:00 – 24:00	null	24h	4
LT-M	07:00 – 24:00	00:01 - 06:59	17h	29

As mentioned, LT - A, B, and C represent the light that the basil is exposed to during the germination stage. That is, the amount of PAR at the soil surface during the 4-day germination period at each location. These three locations (A, B, and C) are important as they are based on practical ease-of-use, light recovery circumstances, and will determine the plausibility of the LT in practice (during normal operation). Figure 7 displays the placement of the separate LT areas and their corresponding phase locations. As directed by the client, LT-A is situated on the floor of the PFAL with its light source being ambient 'waste light' from the module layer. Similarly, LT-B utilizes the same 'waste light' by being placed along the edge of the module cell without obstructing the ordinary operation of the layer. LT-C is placed within separate light shelving that includes 4 x Valoya L-series 14 Watt evenly spaced @ 20cm directly above the pots. Note that the light shelving runs on its own separate time schedule (Table 3).

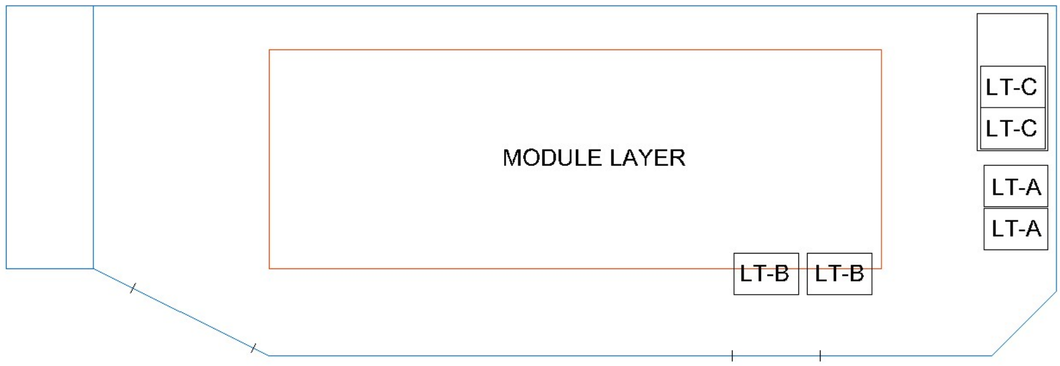


FIGURE 7. PFAL plan view and locations of Light Treatments

The blue outline marks the external walls of the PFAL which are made from a UV-blocking engineered polymer. The red outline marks the extents of the module growing layer which houses the lighting, irrigation, runners, plants etc. LT-A, -B, and -C represent the trays housing the plant pots.

3.2.2 Phase 2

Phase 2 light treatment (LT-M) occurs within the module layer and is somewhat more complex as there is no uniform PAR, at any given height, across all pots within the module. That is, as the basil move through the module layer, during the 29-day phase 2 period, they undergo varying levels of PAR.

TABLE 4. Arrangement of plants and associated LT's

Runner #1	A_{min}	B_{min}	C_{min}	A_{max}	B_{max}	C_{max}	C_{max}	B_{max}	A_{max}	C_{min}	B_{min}	A_{min}
Runner #2	A_{min}	B_{min}	C_{min}	A_{max}	B_{max}	C_{max}	C_{max}	B_{max}	A_{max}	C_{min}	B_{min}	A_{min}

A_{min}, B_{min}, and C_{min} refer to LT's -A, -B, and -C that have been subject to the *minimum* amount of light throughout phase 2. A_{max}, B_{max}, and C_{max} refer to LT's -A, -B, and -C that have been subject to the *maximum* amount of light throughout phase 2. This is done for the simple fact that if a light treatment leads to a plant that is *too large* or *too small* then it will not be marketable and become waste.

Therefore, discovery of the extents to which excessive PAR or insufficient PAR exist will direct the plausibility of the LT.

Determining the DLI across the module is important as it describes the varying light conditions that is experienced by individual plants. The PFAL module LED illuminance level has the ability to be set manually via a control panel inside the PFAL. The LED's have a dimmable function which throttles the output current (%) in integrals of 10 (PARUS LED Grow Light manual 2020). Through experience, the client has set the light output at 70% for the first 4 lamps and 80% for the remainder. Measurements were taken in a grid pattern (Figure 8) at the soil surface level of the peat pots. The vertical distance from the LED's to the top surface of the peat pots is 450mm. The resulting Figure 9 illustrates the varying levels of PAR (µmol/m2s) and DLI (mol/m2d) throughout the module layer. The DeltaOHM HD 2302.0 LightMeter was used to measure the PPFD with the LP 471 PAR Quantum Sensor in the ranges 400nm – 700nm at different points along the module layer.

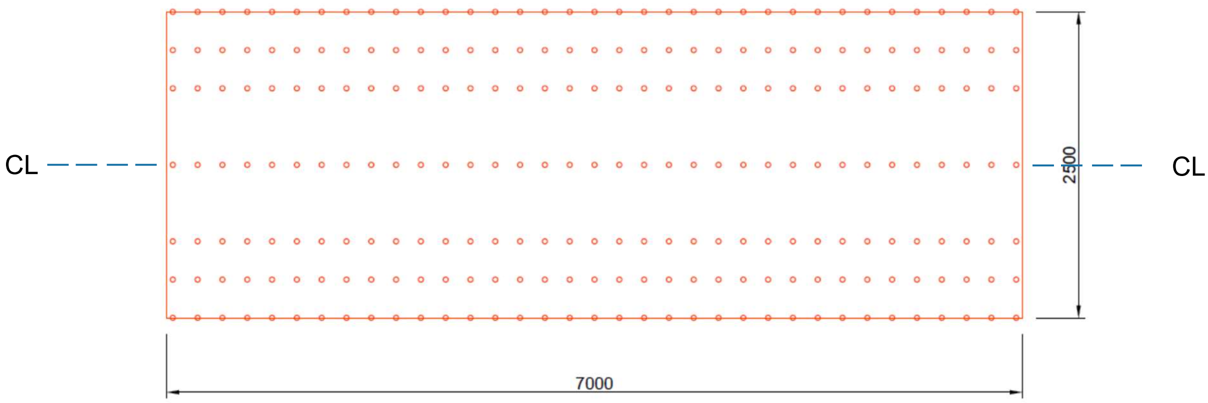


FIGURE 8. Reference grid for mapping PPFD variance across module layer

(The circle marks show where a measure of PPFD has been taken. Dimensions in mm)

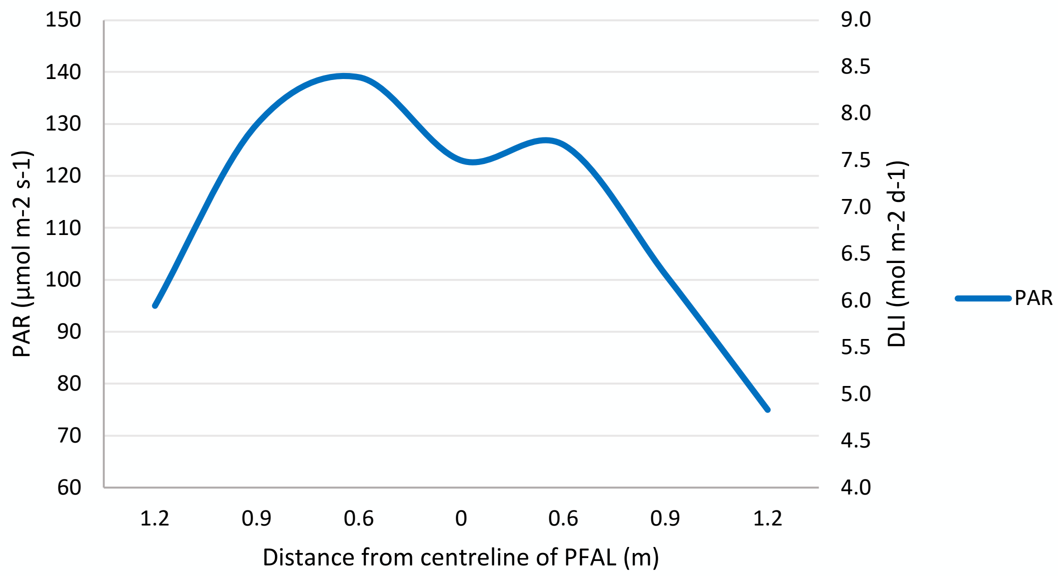


FIGURE 9. PAR variance at soil level across module layer.

The position of the LED's within the layer and the angle of the incident light means that there are several places along the module layer where major variances in PAR can be seen and therefore also experiences by the plants.

3.3. Growth characteristics

On the 33rd day of growth the basil plants were cut approximately 10 mm above the base of the stems. Whole plant Fresh Weights (FW) were measured using an electronic scale (±0.01g) then leaf measurements were made and whole plants were then oven dried for the measurement of Dry Weights (DW) using an electronic scale (±0.01g).

Measurement of the Leaf Area (LA) to determine the Leaf Area Index (LAI) was done manually using vernier callipers, accuracy was made to the nearest 1mm. Measurements were taken of all whole plant samples (n = 23) when harvested in a destructive manner. Sweet basil leaves naturally have a downward curvature both along the midrib and across the width therefore to gain a more accurate measure moistened leaves were pressed into the table surface and spread to their extents as seen in Picture 5. The leaf length (L) along the midrib and width (W) across the widest part, from all plant leaves*, were measured.

For the calculation of LA, Bazaz et al. (2011) formed a regression model to determine Root Square Mean Error (RSME) and coefficients for the dependant variable (LA) against different variables such as L+W, L*W, L/W, L^2+W^2, and L^2*W^2. The best results were an R^2 value of 0.895 with an RMSE value of 0.794 for the model equation:

$$LA = 0.209 * (L^2 + W^2) + 0.25$$

It was concluded that this model could be used to calculate LA, based on L and W measurements, quickly and accurately. This model will be used for the estimation of LA and further to calculate LAI.

$$LAI = LA/ground\ area$$

$$LAI = LA/25$$

For LAI, LA is divided by the ground surface area which was measured as the average top surface area of the pots = 25cm^2.
Basil was oven dried in a Thermo Scientific FunctionLine at 50^{0}C for 14 hours.

PICTURE 6. Left: measurement of harvested basil fresh weight, Right: Measurement of basil leaf length after freezer storage (Denham 2021)

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* Leaves with L< 10mm were not measured

3.3.1 Irrigation

During Phase 1 watering was done manually, using a measured 2 litre container or a pressurized spray bottle. Watering was conducted every day using approximately 2 litres untreated water. During phase 2, watering is done through an automated irrigation system that runs once every 6 hours for 3 minutes dispensing at 41L/hour. Therefore, approximately 8.2L is dispensed per day. The irrigation draws from a water reservoir that was treated with YaraLiva Calcinit EC fertilizer and Kekkilä Superex NPK 8-5-28. Further information about water treatment can be found in Appendix 1. The excess water runs freely and returns to the original reservoir acting as a closed system.

4 RESULTS

A total of 23 plants were measured and the results are given in Table 5. The client made the final decision about the requirements the Sweet Basil must fulfil to be packaged and sent to stores. From the 23 plants used in the experiment 14 were deemed of sufficient size and quality to be used in-stores. Of those 9 rejected plants, 5 were too tall and 4 were too short. 75% of LT-A_{min} were too short and 75% of LT-C_{min} were too tall.

TABLE 5. Results of plant characteristics as per light treatment through manual measurement and calculation.

		Light Treatment					
		A_{min}	B_{min}	C_{min}	A_{max}	B_{max}	C_{max}
Plant characteristic	LA	137±45	211±35	210±37	153±20	224±20	241±20
	FW	10.45±3.97	22.76±6.42	21.41±5.76	15.88±2.93	23.81±1.43	25.35±1.99
	DW	0.87±0.55	1.35±0.42	1.27±0.35	0.91±0.16	1.38±0.04	1.48±0.2
	LAR	13.1±1.6	9.3±1.6	9.8±1.3	9.6±0.8	9.4±0.3	9.5±0.2
	LAI	5.5±1.8	8.4±1.4	8.4±1.5	6.1±0.8	9.0±0.8	9.7±0.8
	Height	21±2	28±1	29±3	24±2	28±2	28±2
	DLI (phs1)	0.75	1.84	7.34	1.06	2.83	9.82
	DLI (phs2)	5.8	7.0	8.0	8.3	8.0	7.7
	n	4	3	4	4	4	4

Plant characteristics in Table 5 are averages of the data set, for example A_{min} LA is an average from n = 4, error margins are therefore included based on standard deviations and a 95% confidence z – score.

Variations in LA corresponding with positions along module layer i.e., variation in phase 2 DLI, show some mixed results. LT-C displays higher LA with higher DLI, and similarly displays lower LA with lower DLI. Increase in DLI at germination shows a major increase in LAI (48 – 53% increase) between LT-A and LT-B cases and only very minor increase in LAI (0 – 7% increase) between LT-B and LT-C.

Average LAI for LT-A was a minimum $LAI_{av.Amin}$ = 5.5 and maximum $LAI_{av.Amax}$ = 6.1, due clearly to the lower overall DLI in phase 1 (0.75 – 1.06 μmol m^{-2} s^{-1}), compared with LT -B and LT -C. $LAI_{av.Amin}$ showed a 35% reduction in LAI as compared with $LAI_{av.Bmin}$ and $LAI_{av.Cmin}$. $LAI_{av.Amax}$ showed a 32% and 37% reduction in LAI as compared with LT $-B_{max}$ and LT $-C_{max}$ respectively. Figure 10 shows that LAI and FW followed a relationship with an R^2 value of 0.92, whereas LAI and DW followed a relationship with an R^2 value of 0.69. In both relationships, FW and DW increase linearly with an increase in LAI.

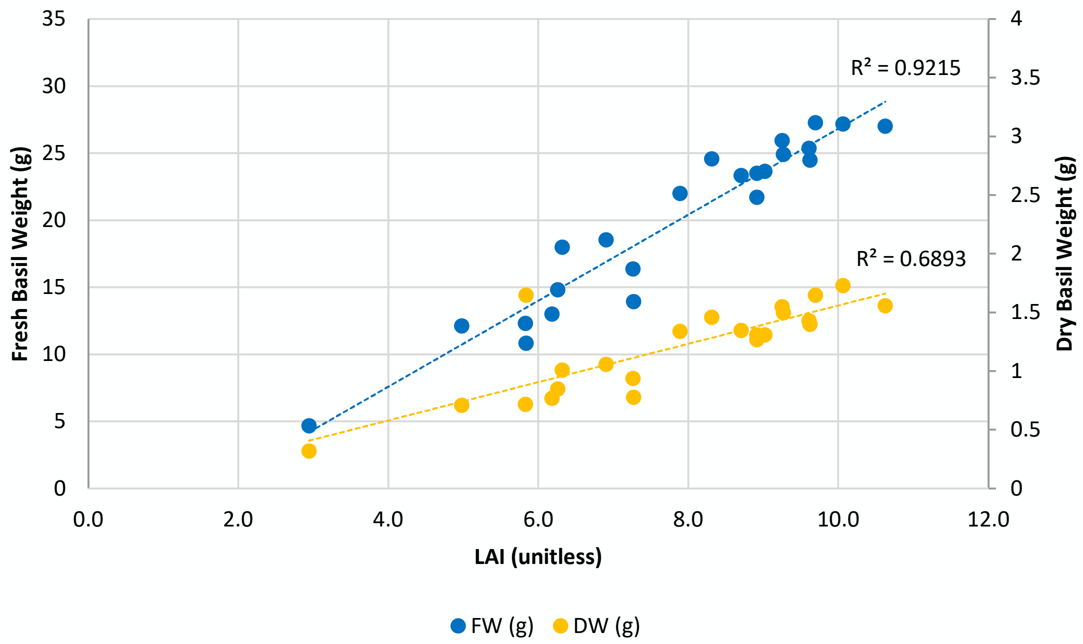

FIGURE 10. Fresh and Dry Weights vs. LAI of all 23 basil plants at 33 days growth period.

Height of the basil was compared with FW and LAI (Figure 11) and shows good correlation with an R^2 value of 0.83 and 0.8, respectively. With both LAI and Height having strong correlation to FW it can be noted that stem length increases linearly with leaf area. That is to say, morphologically, energy is being transmitted into the vertical growth (increase in stem length, production of more nodes) directly proportional to the horizontal growth (leaf area expansion).

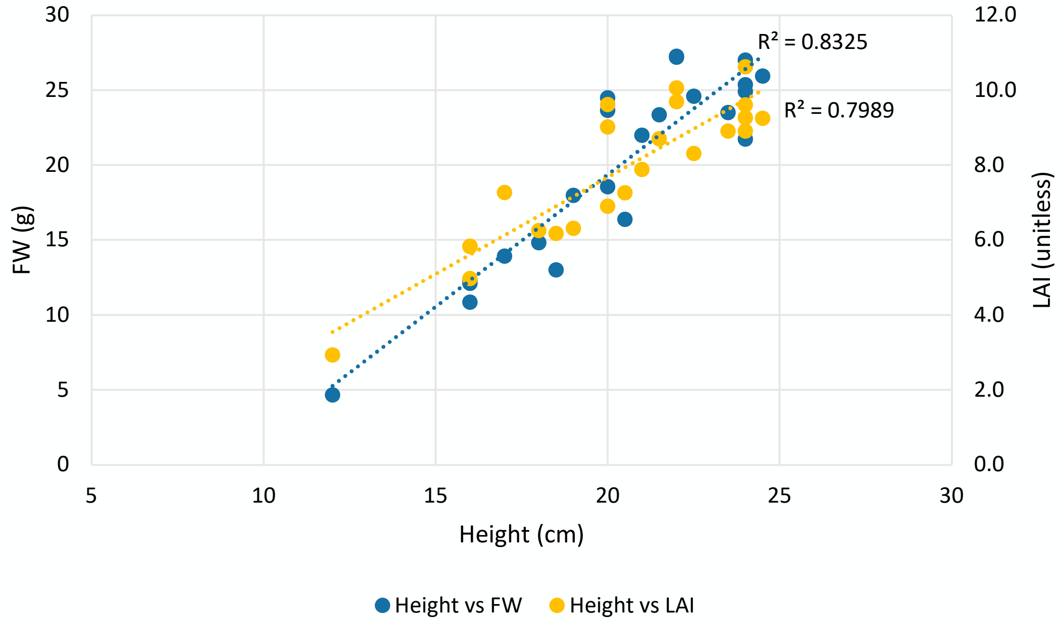

FIGURE 11. Height vs. Fresh Weight and Leaf Area Index correlations

Average leaf area ratios (Figure 12) were 31% higher for the lowest light treatment DLI = 0.8mol m^{-2} d^{-1} (LT-A_{min}). Other light treatments showed statistically insignificant differences.

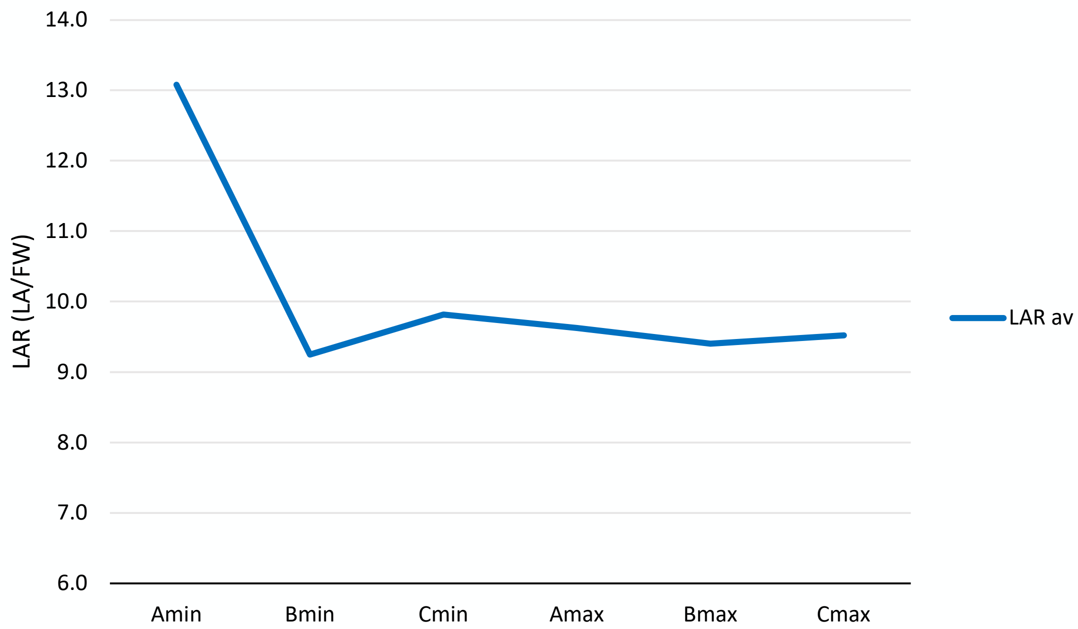

FIGURE 12. Average Leaf Area Ratio (LA/FW) per Light Treatment.

LAI regressed against FLI (Figure 13) displays a shallow exponential growth beyond an LAI of approximately 3. The regression lies most entirely within the fraction light intercepted range between 0.8 and 1.0, meaning the crop canopy is mostly closed.

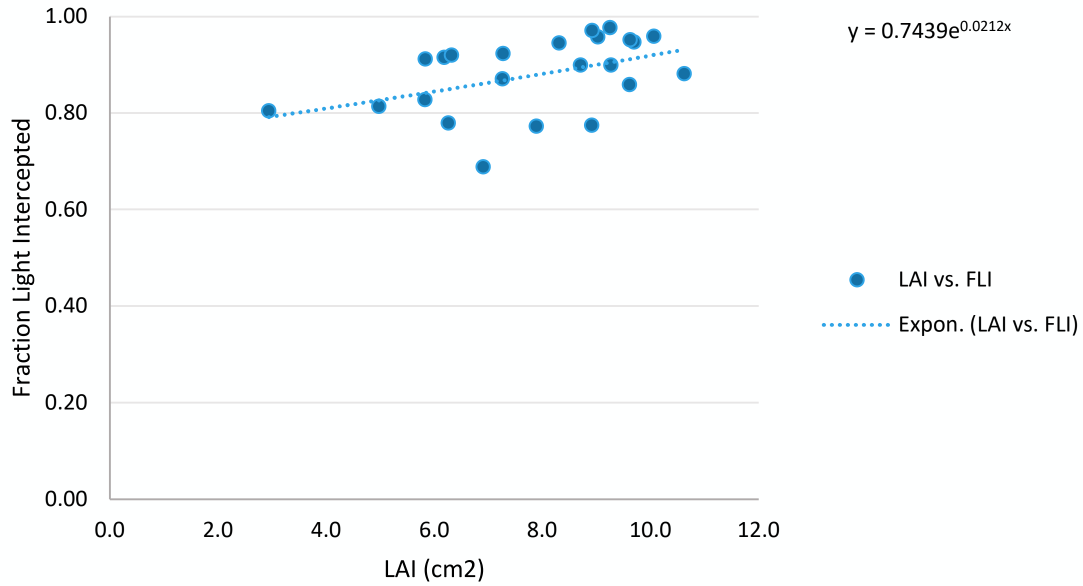

FIGURE 13. Leaf Area Index vs. Fraction Light Intercepted

In Figure 14, A, B, and C correspond to average LAI values per Light Treatments -A, -B, and -C. The two similar coloured points mark the min and max LT's. In Figure 14 a), across all LT's a logarithmic trendline can be made i.e., y = 1.3882ln(x) + 6.5185 (not shown). Phase 1 DLI displays much higher variation between LT's when compared with phase 2 DLI.

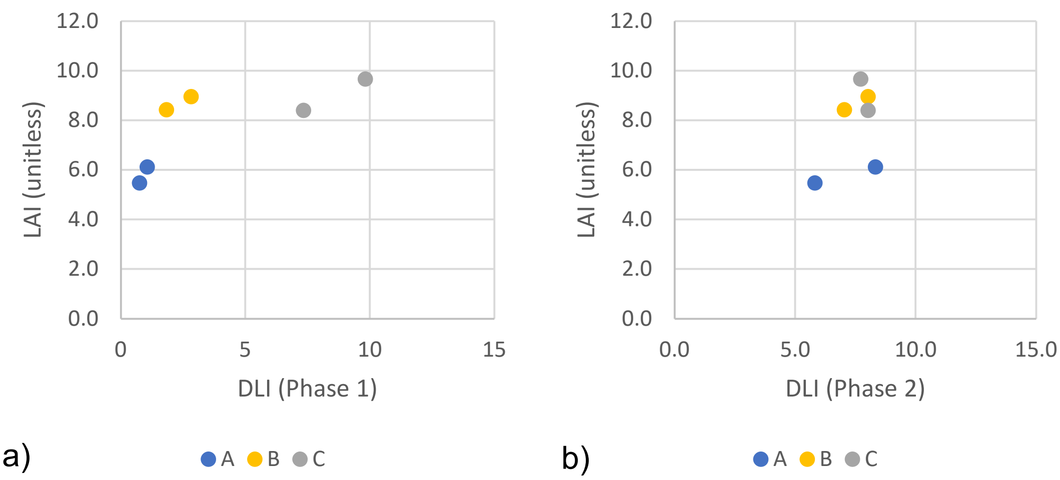

FIGURE 14. **a)** Phase 1 Daily Light Integral vs Leaf Area Index, **b)** Phase 2 Daily Light Integral vs Leaf Area Index

4.1. Climate results

Regression models were made for temp, RH% and CO2 levels monitored vs. manual. Air temperature, RH%, and CO2 returned RSME values of 0.87, 9.95, and 22.08 respectively. This provides good confidence in the monitoring measurements of air temperature and to a lesser extent CO2, however less confidence in RH%.

5 DISCUSSION

At The Little Garden ® it is not a necessity that this variety of sweet basil is produced here, or for that matter basil at all, however there has been a significant investment in the design of the system to accommodate many different kinds of herb and sprout species so that it is marketable as a versatile PFAL system, therefore a commonly used greenhouse herb must be a suitable option. With this in mind, it was found that the selected light treatments as well as the variation in PAR within the module effected the growth of the Sweet Basil in several ways and some of the treatments were plausible alternatives during germination.

5.1. Climate

Average air temperature throughout major growth (LT-M) was within typical optimum temperatures for plant growth. Average CO_2 level at 510ppm could benefit from higher levels as an increase in lower ranges of CO_2 (such as the 400-500 ppm range) has a large effect on photosynthesis rate. (Heuvelink et al. 2019, 151-155.) 77% average Relative Humidity rate is within typical optimum ranges (70 – 90%).

5.2. Plant characteristics

LAI across all treatments were very high with 95% of plants having LAI > 4. One explanation may be that there is a small ground surface area as provided by the pots i.e., ~25cm^2. As expressed by Kozai et al. (2016, 147-148), LAI has marginal effect on canopy photosynthesis beyond an LAI of 4 (Figure 2) since most of the incident radiation is absorbed by the canopy leaves. The regression of LAI and FW, however, show high confidence of their relationship with R^2 = 0.92. This shows that LAI > 4 provides a linear increase in FW, therefore gains from photosynthesis are higher than the losses due to respiration. For this reason, it is not strictly necessary to limit the amount of LAI for the fear of loss in efficiency. This relationship also shows that LAI may be a good prediction of FW in this model regardless of light treatment. The regression of LAI and DW show less confidence with $R^2 = 0.69$. Leaf thickness and assimilates thickness may be an explanation here, however these measurements were not taken.

Figure 6 from Heuvelink (2005) shows the exponential function of LAI versus FLI for tomato leaves and how an increasing canopy promotes an inverse exponential growth of FLI, where an asymptote is reached around an LAI of 4. Beyond an LAI of 4 the increase in FLI is expected to flatten out between 90 and 100%. In this experiment at 33 days growth the basil LAI versus FLI shows a slow upwards slope $y = 0.7439e^{0.0212x}$ (Figure 13), indicating a shallow exponential increase. Only one value in the data set lies below an LAI of 4, therefore as compared with tomato leaves, this shallow slope is a fair expectation albeit a small margin outside of the range with 80 – 100% FLI. Additionally, the exponential equation for basil in this case is not an inverse one which may be due to the lack of range of data below LAI of 4. Another cause is that tomato leaves and basil leaves have morphological differences in how horizontal they are in space and their thickness, allowing different diffusal rates.

Average LAI and Phase 1 DLI correlation (Figure 14 **a)**) shows a relationship between maximum and minimum irradiance and across LT's. Lower DLI corresponded to lower LAI for LT -A and LT -B, however the opposite was true for LT-C. It is clear that the range of DLI per Light Treatment effects the growth of the basil only up until a limiting point wherein the capacity of the plant to increase leaf expansion is increasingly *less* dependant on the intercepted light at the germination phase (Phase 1). That is, a logarithmic trend is present where $DLI \to \infty$. The substantial increase in DLI between LT-B to LT-C (Figure 14 **a)**) and the corresponding minimal expansion in LA (Figure 14 **b)**) suggests that LT-B is sufficient in PAR over the other LT's. It might also be considered, as detailed by Kozai et al. (2016, 147), that the expansion in LA beyond LAI > 4 has increasingly trivial importance on canopy photosynthesis as there is now a closed canopy. The minor difference in LAI between LT-B and LT-C corresponding to Phase 2 DLI re-enforces this. In contrast Beaman et al. (2009) showed that sweet basil (and two other cultivars) had a saturated photosynthetic capacity of ~500µmol m^{-2} s^{-1} with a 16h photoperiod. This means a DLI of ~28.8mol m^{-2} d^{-1} was possible for maximum biomass production rate, which is almost 3 times the maximum DLI at germination (9.8mol m^{-2} d^{-1}) and 4 times the average maximum DLI across the module (7.2mol m^{-2} d^{-1}) in this experiment. The low DLI values in The Little Garden may still be effecting the total growth rate possibilities, and should be re-evaluated, however other factors are possibly causing low growth rates for example water, CO_2, or heat stress. Further investigation is needed here.

Leaf Area Ratio is describing the partitioning of energy into either the expansion of the leaves for photosynthesis or growth of the plant biomass i.e., the overall leafiness of the plant (Hunt 1990). There is a clear relationship between the average LAR across different light treatments. Near equivalent LAR's can be seen for all light treatments, with the exception of LT-A_{min}. 38% higher average LAR for LT-A_{min} means that an increasing LA was producing less FW than the other light treatments. This may be because the lower DLI across all LT-A_{min} plants was causing growth to be more focused on leaf expansion for increased light interception rather than on assimilation such as stem growth and biomass in general. This would suggest that for LT-A_{min} Height vs. LAI a poor regression model would be found since LA would be increasing at a higher rate over increases in height as compared with other LT's, which is not the case (R^2 = 0.8). Further investigation is needed into the determinates of the plant biomass production, for example correlations between internodal length, leaf thickness, stem thickness, number of stems, and others.

5.3. Germination seed count

The method of seed dispersal has obvious limitations and a high degree of variation between seed counts per pot, however there are some factors which are relevant here. Firstly, there are financial limitations that drive many aspects of the operation and so cheap alternatives are required. Secondly, the success of individual seeds in becoming an adult plant is driven, in no minor way, by the available space for root growth and canopy area above the soil, similarly and in contrast the failure of germination and growth is attributed to the *lack* of such space, therefore the sowing as per this method, wherein an excess of seeds are sown per pot, limits the number of adults plants that can succeed regardless (to a degree) of the seed count. In short, per soil area there is a limiting number of successful plants that will reach lengths up to or greater than desired, and so an excess of seeds are sown per pot.

5.4. Possible errors

During harvest one plant from Light Treatment batch B_{min} was misplaced, therefore only 3 samples from this light treatment were able to be measured. This causes an anomaly in the sample sizes possibly effecting the plant characteristic averages.

Time and financial restraints prevented the monitoring of a larger data set during the time-period. The methods used were destructive in nature and therefore any basil used in the experiments were unable to be sold afterwards. Therefore, only a set number of pots from each batch could be used in the experiment as the rest were needed for the normal operation of the business to continue. A random selection process for those plant pots to be selected and arranged within the module, along with a higher number of monitored pots, would provide a more reliable data set for estimating basil variance across LT's AND across batches.

Measurement of the leaf lengths and widths was time consuming (there were just under 2000 hand measurements made) and therefore data collection needed to be made over an extended period, which had not been planned for. Basil plants needed to be then packed and frozen before measurements could be made, this was to prevent degrading of the leaf and impact of the leaf dimensions. This method however made the basil very moist and had possibly impacted the true size of the leaves when measured.

The reference photos that were being made approximately every two weeks required that the basil plants be removed from the module layer manually at different stages during the growth period. This may have impacted on the DLI, although the time outside of the module was minimal therefore the impact is considered negligible.

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7 APPENDICES

7.1. APPENDIX 1. Chemicals

YaraLiva Calcinit EC Fertilizer - Calcium Nitrate:

Element	g/Kg
Total Nitrogen	155
Nitric Nitrogen	144
Ammoniacal Nitrogen	11
Calcium	190
Calcium Oxide	265

Kekkilä Superex NPK 8-5-28 (Mg 2.5):

Superex fertilizer as it was supplied in powder form was incorporated into the irrigation system at 1.412Kg fertilizer per 1000L solution. The table below details the micro and macro nutrients within the solution in granular form grams element per Kilogram superex and in solution form milligrams element per litre solution.

Element	g/Kg	mg/L
N	100	141
P	40	56
K	300	424
Ca	-	-
Mg	14	20
S	19	27
	mg/Kg	mg/L
Fe	1950	2.75
Mn	700	0.99
B	340	9.48
Zn	170	0.24
Cu	90	0.13
Mo	51	0.072
Cl		
Conductivity mS/cm		1.55

For the Calcium Nitrate content (832g/1000L):

Element	g/Kg	mg/L
N	155	129
Ca	190	158