

The Assessment of Growth Performance of Brassica rapa var. chinensis 'Li Ren Choi', Spinacia oleracea 'Auroch', Eruca sativa 'Astro', and Brassica rapa var. japonica Using GREENBOX Technology

George Paul Buss¹, Paige Ann Carroll², Mya Alexandria Catherine Griffith^{1,2}, Xiusheng Yang³, John L. Griffis Jr.², Galen Papkov⁴, Sarah Bauer⁵, Kathryn Jackson⁶, Ankit Kumar Singh² [®]

¹Department of Marine and Earth Sciences, The Water School, Florida Gulf Coast University, Fort Myers, FL, USA

²Department of Ecology and Environmental Studies, The Water School, Florida Gulf Coast University, Fort Myers, FL, USA

³Department of Natural Resources, University of Connecticut, Storrs, CT, USA

⁴Department of Mathematics, Florida Gulf Coast University, Fort Myers, FL, USA

⁵Department of Environmental and Civil Engineering, Mercer University, Macon, GA, USA

⁶Independent Researcher, Alexandria, VA, USA

Email: gbuss@fgcu.edu, pcarroll@fgcu.edu, mgriffith@fgcu.edu, xiusheng.yang@uconn.edu, jgriffis@fgcu.edu, gpapkov@fgcu.edu, bauer_sk@mercer.edu, kgn816@gmail.com, asingh@fgcu.edu

How to cite this paper: Buss, G.P., Carroll, P.A., Griffith, M.A.C., Yang, X.S., Griffis Jr., J.L., Papkov, G., Bauer, S., Jackson, K. and Singh, A.K. (2023) The Assessment of Growth Performance of *Brassica rapa var. chinensis* 'Li Ren Choi', *Spinacia oleracea* 'Auroch', *Eruca sativa* 'Astro', and *Brassica rapa var. japonica* Using GREENBOX Technology. *Agricultural Sciences*, **14**, 1222-1237. https://doi.org/10.4236/as.2023.149082

Received: May 27, 2023 Accepted: September 1, 2023 Published: September 4, 2023

Copyright © 2023 by author(s) and Scientific Research Publishing Inc. This work is licensed under the Creative Commons Attribution International License (CC BY 4.0).

http://creativecommons.org/licenses/by/4.0/

Abstract

Obtaining nutritious food is becoming increasingly difficult due to the growing urban population and the degradation of soil, water, and air from mechanized and industrialized agricultural techniques. More than half the global population resides in urban areas, with not enough surrounding agricultural land to meet food requirements. Food traveling long distances, an average of 1020 miles, has resulted in increased food miles for the average food item in the United States of America, representing wasted resources. The novel GREENBOX technology was invented in response to increasing pressures on food security. Previous studies conducted on GREENBOX technology assessed the technical feasibility of utilizing Lettuce Lactuca sativa 'Rex Butterhead'. We at the APS Laboratory for Sustainable Food at Florida Gulf Coast University assessed the technical feasibility of growing different leafy green vegetable crops. GREENBOX technology consists of thermally insulated climate-controlled enclosures, an artificial lighting source, a soilless cultivation method (hydroponics), and environmental control modules. We assembled two GREENBOX units to assess the environmental conditions and growth performance of Brassica rapa var. chinensis 'Li Ren Choi', Spinach Spinacia oleracea 'Auroch', Arugula Eruca sativa 'Astro', and Mizuna Brassica Brassica

rapa var. japonica. Plugs were cultivated and then transplanted in a randomized manner to the nutrient film technique (NFT) channels, subsequently grown for 30 days to full bloom and ready for harvest. Fertigation was carried out using a standard concentration nutrient solution. Crops were arranged in twelve blocks of four species each. We collected environmental data including daily light integral (DLI, mol/m²·d), temperature (°C), relative humidity (%), and vapor pressure deficit (VPD, kPa). Collected biomass data included wet weight (g), dry weight (g), leaf area (cm²), and chlorophyll concentration (mg/cm²). We then derived the Specific Leaf Area (SLA, cm²/g). Descriptive statistics were utilized to understand the differences in biomass parameters between the four crops grown. We also compared the performance parameters of our crops with existing peer-reviewed literature and found it superior, if not comparable to commonly found industrial output. We determined that all crops grew to full bloom, demonstrating that GREENBOX technology may be used to grow a variety of different leafy green vegetable crops.

Keywords

Controlled Environment Agriculture, Food Insecurity, GREENBOX, Hydroponics, Lettuce

1. Introduction

The security of food production systems is under increasing pressure from a variety of factors as discussed below. The global population has increased from 2.5 billion in 1950 to a projected population of 11.2 billion nearing 2100 [1]. A result of global population growth is an unprecedented increase of pressure on food production systems [2]. As the population grows, it does not spread evenly and is more concentrated in urban areas. According to the United Nations, 2007 marked the first time in history the number of residents in urban areas was greater than that in rural areas, and urban populations are expected to continue increasing to over 60% by 2030 [3] [4]. There are increased pressures on global food security [5] as the two major agricultural powers in war, Russia and Ukraine, both provide crucial agricultural products to global markets [6].

With more residents in urban areas than ever before, food deserts are becoming more prominent accompanied by rising food insecurity. Food deserts are identified as urban areas with lower accessibility to fresh foods which primarily affect socio-economically disadvantaged people [7]. Food insecurity is a term that describes the phenomenon of people in food deserts having limited access to adequate, safe, and nutritious food for daily needs and healthy living [8] [9]. Major chain supermarkets are oftentimes unwilling to locate their stores in the inner city or low-income neighborhoods due to obstacles such as lower profitability from high overhead costs and low profit margins, increased crime rates, and cultural biases [10] [11] [12] [13]. The absence of supermarkets and fresh produce in food deserts allow an abundance of readily available calorie-dense foods that are nutrient poor to be present in food deserts [14]. A nutrient rich balanced diet is essential to human health and since these foods cannot be easily obtained by people living in food deserts, they are more likely to develop obesity and diabetes, which together account for \$395 billion in annual medical costs and lost productivity [15] [16]. Decreased security of food production systems is the result of urban population growth and food deserts. As of 2018, 11% of the United States population faced food insecurity and this number continues to increase [17].

Traditional soil-based agriculture requires an immense amount of space for efficient crop production, currently encompassing an area of 48 million km² [18]. With industrial farming moving farther from urban areas, food must be transported longer distances to reach consumers. The number of miles food must travel from its origin to consumer is termed food miles [3]. On average, food travels over 1020 miles (1640 kilometers) from source to consumer in the United States of America [3].

According to the Food and Agricultural Organization (FAO), meeting the demand for a rapidly growing population requires a 70% increase in food production capacity between 2005 and 2050, despite the declining arable land per capita [19]. Although immense increases in food productivity are necessary, there are many threats to conventional agriculture which include soil, air, and water pollution, soil salinization from excessive fertilizer use, desertification, climate change induced droughts, extreme variation in temperatures, extreme variation in solar radiation, and the spread of pests [20]. As natural resources deplete, the current global agricultural system cannot be sustained, and food production demands cannot be met, creating a need for a more resource efficient avenue of fresh food production.

Controlled environment agriculture (CEA) may serve as an alternative avenue for crop production. The term, controlled environment agriculture, was first conceptualized in the 1960s, but had minuscule applications in the agricultural industry [21] [22]. CEA is defined as a closed-system farming method that uses a smaller footprint to grow crops [23]. The six main features commonly found in CEA setups include: 1) a structure that is thermally insulated with non-transparent walls; 2) a multi-tiered system that accommodates crops and lighting; 3) pumps to remove heat generated by lights and dehumidify the unit; 4) carbon dioxide delivery unit to enhance photosynthesis; 5) nutrient delivery system; and 6) environmental control units for electrical conductivity and pH regulation to support nutrient flows [24]. CEA commonly uses soilless cultivation systems such as hydroponics, aeroponics, or aquaponics [25].

GREENBOX technology was developed using principles of CEA by the Yang Laboratory at the University of Connecticut using novel low-cost independently functioning units capable of fresh crop production in warehouse settings [26]. Technical feasibility studies were carried out [27] [28] [29] and found GREENBOX technology to be technically viable for crop production year-round. The performance was comparable to greenhouse production [30] [31] and found to be fi-

nancially feasible, costing \$400 a unit [32].

Previous studies on crop production using GREENBOX technology grew Lettuce *Lactuca sativa* 'Rex Butterhead'. We at the APS Laboratory for Sustainable Food at Florida Gulf Coast University were interested in determining the technical performance of different leafy vegetable crops using GREENBOX technology. The main aim of this experiment was to assess the environmental conditions and biomass performance of *Brassica rapa var. chinensis* 'Li Ren Choi', Spinach *Spinacia oleracea* 'Auroch', Arugula *Eruca sativa* 'Astro', and Mizuna Brassica *Brassica rapa var. japonica*. Results from this study will help expand the scope of applicability using GREENBOX technology for different types of crop production.

2. Materials and Methods

2.1. Location

The experiments were carried out at Florida Gulf Coast University (FGCU) in the Aquarium Room 114 of Academic Building 9. The experiment location was maintained between 20.5°C and 22.2°C by the Work Management Center. As with previously conducted studies on GREENBOX technology at the University of Connecticut, the laboratory had warehouse-like conditions defined by tall ceilings and fewer windows. The climate of Southwest Florida is characterized by a tropical climate, with a wet summer season and a contrasting dry season from mid-fall to late spring [33]. The average annual temperature in Fort Myers was 24.5°C with a low temperature of 18.8°C in the month of January and a high temperature of 28.7°C in the month of August [34].

2.2. Experimental Setup

Our experimental setup was similar to previously conducted studies of GREENBOX technology [27] [28] [29], which included two thermally insulated grow tents, light emitting diodes (LED) lighting elements, a nutrient film technique (NFT) hydroponic system, a nutrient reservoir, and environmental control modules.

We assembled two thermally insulated grow tents (The Original Gorilla Grow Tent* 5 × 5, Gorilla Inc., Santa Rosa, California) which had dimensions of $1.52 \times 1.52 \times 2.12$ m and weighed 33.9 kg. Constructed with dense 1680D fabric material, each tent was free of disturbance from external influences allowing for a uniform controlled environment throughout the growth process.

To facilitate photosynthesis for our chosen crops, we installed four light fixtures (FREELICHT 4 ft LED Grow Light 60 W, Amazon, Seattle, Washington) in each GREENBOX unit. Each light fixture had three LED bead colors of white, yellow, and red. Lights were attached to the ceiling of the tent by support ratchets (Heavy-Duty Stainless-Steel Gear Ratchets, AC Infinity, Los Angeles, California) which collectively produced 12,000 lumens of light and 14,000 K color temperature. We used a programmable timer (Mechanical 24-Hour Programmable Dual Outlet Timer, BN Link, Santa Fe, California) with LED light sources to control light duration throughout the entire growth process. The lights were positioned 0.31 m above the crop canopy.

Two Fans (CLOUDLINE T6, 6" Inline Duct Fan with Temperature Humidity Controller, AC Infinity, Los Angeles, California) were utilized to establish a ventilation system in the two GREENBOX units. The fans had duct openings that measured 0.15 m in diameter, an airflow capacity of 11.38 cubic meters per minute (CMM), and a power rating of 38 watts. A single fan was placed in the top vent of each GREENBOX unit in a position that directed airflow outwards. The bottom vent was left open to provide air intake, creating recirculating air movement in the tents. The built-in environmental sensors were placed in the center of the grow tents at the same elevation as the crops and continuously recorded humidity, temperature, and vapor pressure deficit every 15 minutes throughout the growth cycle which represented critical environmental conditions the crops were exposed to [35]. We utilized light sensors (FUTUREHORTI Light PAR Meter PPFD Tester, Amazon Inc., Seattle, Washington) to collect data on the photosynthetic photon flux density (PPFD, μ mol/m²·s) and daily light integral (DLI, mol/m²·d). Monitoring the DLI of each GREENBOX unit allowed us to ensure plants were receiving sufficient light for growth.

The soilless cultivation system in each GREENBOX unit consisted of four Nutrient Film Technique (NFT) hydroponic channels (HydroCycle 4" Pro NFT Series 4' Channels, FarmTek^{*}, Dyersville, Iowa) that were uniformly placed 0.3 m apart on a tray stand (SKU number HGC706122, Fast Fit Ltd., Hawthorne Gardening Company, Vancouver, Washington) with 1.22 × 1.22 m dimensions. Each channel was made with UV stabilized plastic and had dimensions of 0.10 × 0.05×1.22 m. Each NFT channel had six square holes that were 0.15 m apart where the crops were placed.

We measured the pH and electrical conductivity using a portable pH/EC meter (CANNABMALL TDS PPM pH Meter, Amazon Inc., Seattle, Washington) to ensure the nutrient solution stayed within the desired pH and EC range. The nutrient solution was placed in a reservoir (Hudson Exchange High Density Polyethylene 7 Gallon Bucket, Amazon Inc., Seattle, Washington).

A submersible pump (Active Aqua AAPW400 Submersible Water Pump, Hydrofarm^{*}, Petaluma, California) was placed in the nutrient solution reservoirs. With a power rating of 25 W and 120 V, the pump circulated 25 L of nutrient solution per minute into the NFT channels providing the crops with the necessary water and nutrient requirements for growth. The NFT channels were positioned with a slight incline allowing gravity to carry the nutrient solution from the inflow to the outflow tubing and back into the nutrient reservoir, establishing a closed system of nutrient solution flow. The nutrient solution to support crop growth was formulated by utilizing a combination of (Jack's Nutrients 5-12-26 Part A FeED, JR Peters Inc., Allentown, Pennsylvania) and (Jack's Nutrients 15-0-0 Calcium Nitrate Part B, JR Peters Inc., Allentown, Pennsylvania).

We used four different crops in this experiment which included *Brassica rapa* var. chinensis 'Li Ren Choi', Spinach Spinacia oleracea 'Auroch', Arugula Eruca

sativa 'Astro', and Mizuna Brassica *Brassica rapa var. japonica*, all grown from seeds (Johnny's Selected Seeds, Fairfield, Maine). We chose these leafy green vegetables because they possess favorable characteristics for growth in controlled environments which includes a maximum height of roughly thirty centimeters and a relatively short growth cycle between ten to thirty days [36] [37]. Figure 1 illustrates the experimental setup.



Figure 1. The experimental setup for crop production of *Brassica rapa var. chinensis* 'Li Ren Choi', Spinach *Spinacia oleracea* 'Auroch', Arugula *Eruca sativa* 'Astro', and Mizuna Brassica *Brassica rapa var. japonica* using GREENBOX technology.

2.3. Experimental Procedure

We conducted our experiments between February and April 2023 which included a plug preparation stage followed by a 30-day growing period. We first began by sowing the four different crop seeds in a starter growth medium (Horticube XL 104-Cell Sheets, Oasis[®] Grower Solutions, Kent, Ohio) with dimensions of $2.54 \times 3.18 \times 3.81$ cm. After saturating the Horticube with plain reverse osmosis (RO) water, we placed a single seed in each cell of the Horticube, totaling 27 of each crop. We covered the seeds with a layer of newspaper and put the covered tray inside the GREENBOX unit in complete darkness for 48 hours.

After uncovering, LED lighting elements were programmed to provide 16 hours of light per day from 06:00 to 22:00 which remained consistent for the rest of the growth cycle. We fertigated the seeds with a starter synthetic fertilizer solution which was formulated by mixing 3.6 grams of "Jack's Nutrients Hydroponic 15-0-0" (calcium nitrate) and 3.8 grams of "Jack's Nutrients Part A 5-12-26" in every 10 L of RO water. We irrigated the seedling tray by adding 100 - 400 mL of starter solution daily to maintain saturation of the Horticube until two leaves after the cotyledons had developed for each crop type indicating they were ready for transplant into the NFT channels.

Once plugs were prepared and ready for transplantation, we randomly selected 12 plugs of each plant species to be placed in each GREENBOX unit, giving preference to healthier looking plugs. In a randomized manner, we then placed one of each crop species per row of the GREENBOX units and began fertigation. The nutrient solution utilized in the GREENBOX unit was formulated by mixing 9 grams of "Jack's Nutrients Hydroponic 15-0-0" (calcium nitrate) and 9.4 grams of "Jack's Nutrients Part A 5-12-26" for every 10 L of RO water.

We checked the pH and EC of the nutrient solution every five days. We ensured the nutrient solution stayed within the desired pH range of 5.8 ± 0.2 Standard Units (SU) and EC range of 1.5 to 2.0 mS. If the pH fell below the desired range, we added 0.5 M NaOH solution using a pipette until the pH rose to 5.8 SU. Similarly, if the pH rose above the desired range, we added 0.5 M HCl solution to the nutrient reservoir until the pH lowered back to the desired range. To maintain the EC range of 1.5 to 2.0 mS, we added fertilizer solution when the EC was lower than desired and added RO water to dilute the nutrient reservoir when the EC was greater than 2.0 mS. Crops were monitored daily for 30 days until they reached full maturity. We did not utilize Integrated Pest Management (IPM) techniques during the growth cycle as our crops did not exhibit signs of disease or pest infestation, however, bio-controls may be utilized [27].

2.4. Data Acquisition

To collect environmental data, we utilized the built-in environmental sensor of the fans (CLOUDLINE T6, 6" Inline Duct Fan with Temperature Humidity Controller, AC Infinity, Los Angeles, California). The sensor of this fan recorded the temperature, relative humidity, and vapor pressure deficit (VPD) every 15 minutes of the growth cycle. To collect light data, we utilized light sensors (FUTUREHORTI Light PAR Meter PPFD Tester, Amazon Inc., Seattle, Washington) every sampling event to determine the photosynthetic photon flux density (PPFD) and daily light integral (DLI) the crops were exposed to in the growth cycle. Monitoring the DLI of each GREENBOX unit allowed us to ensure plants were getting sufficient light for growth.

To evaluate the growth performance of each crop species, we collected biomass data every 5 days of the growth period by harvesting one of each crop species per GREENBOX unit. On the 30th day, we harvested the remaining crops and described the growth trends for each crop.

For each sampling event, we utilized numerous destructive methods to obtain biomass data which included total leaf area (cm²), wet weight (g), dry weight (g), and total chlorophyll content (mg/cm²). Wet weight was found by gently pulling the crop from the NFT channel, separating the remaining Horticube material and root material, and weighing the samples. Separation of root and Horticube material was performed before each weighing event because the root mass contained varying amounts of saturated Horticube material, which if included in the weight, would lead to an inconsistent analysis of the biomass. Wet weight was measured first and immediately after harvest for each crop to ensure minimum water loss through evapotranspiration, which would dry out the crop samples and lead to inaccurate measurements that are lower than the actual value.

After wet weight was obtained, we utilized a chlorophyll meter (atLEAF CHL Plus Chlorophyll Meter, FT Green LLC, Wilmington, Delaware) to find the total chlorophyll content of each crop sample. We took four readings per crop specimen and after averaging the four values obtained, converted the average value to SPAD (Soil Plant Analysis Development) units and subsequently to total chlorophyll content (mg/cm²). Total chlorophyll content values were calculated by converting the atLEAF CHL values to SPAD units. Once converted to SPAD units the relationship between chlorophyll content and SPAD units is used to find the total chlorophyll content for each crop species [38].

To calculate the total leaf area for each plant, we installed the Leafscan app on a mobile device (iPhone 11, Apple Inc., Cupertino, California). We separated each leaf of the crop and laid them out on a white sheet with a 10.5×10.5 cm reference square. Utilizing the camera on the mobile device to take a picture, the Leafscan application ran an algorithm that measures the green leaf area in comparison to the blank white area, thus generating a total leaf area value [39]. The Leafscan app calculated the area inside the leaf contours in pixels, and by using the 10.5 cm reference square, converted the leaf pixel area into the total surface area [39]. We collected the total leaf area of each GREENBOX unit crop sample and exported the area values in a comma-separated values (csv) format. The Leafscan app measured the leaf area in square centimeters (cm²) with an accuracy of 0.01 cm² [39].

The dry weight of each crop was calculated by placing the sample in an indi-

vidually labeled brown paper bag. The paper bags containing the different crops were then placed in a drying oven set at a temperature of 65°C for six days. After six days in the oven, we returned to weigh the completely dried crop samples and recorded their weights.

2.5. Data Processing and Statistical Analysis

We processed the collected environmental data as described in the previous section to determine the environmental conditions the crops were exposed to throughout the growth cycle. By utilizing the built-in environmental sensor of the fan component in each GREENBOX unit, we obtained the relative humidity, temperature, and VPD every 15 minutes of the growth cycle. We exported these values in a csv format. We then derived the average, minimum, and maximum values of the three critical environmental parameters which allowed us to evaluate the ideal environmental conditions for crop production.

Utilizing data collected throughout the growth cycle, we derived the specific leaf area (SLA, cm²/g) and total chlorophyll content (mg/cm²). Specific leaf area is derived from considering the ratio of the total leaf area (cm²) to the dry weight (g) for each sample. To find the total chlorophyll content (mg/cm²) of each different crop, we utilized a chlorophyll meter (atLEAF CHL Plus Chlorophyll Meter, FT Green LLC, Wilmington, Delaware). The atLEAF chlorophyll meter determines the transmittance of light through the leaf surface in wavelengths (660 to 940 nm) associated with chlorophyll to find a value [38]. By employing use of the calculations performed by Zhu *et al.* [38], we found that SPAD values (r² = 0.78) and the values computed by the atLEAF chlorophyll meter (r² = 0.72) have a strong correlation. The correlation between SPAD and atLEAF values was used to convert the computed atLEAF value to the corresponding SPAD unit value which was then converted to total chlorophyll content (mg/cm²) using the formula ($y = 5.52E-04 + 4.04E-04x + 1.25E-05x^2$) described by Richardson *et al.* [40].

We compared the biomass output of each crop species to previous studies to determine if *Brassica rapa var. chinensis* 'Li Ren Choi', Spinach *Spinacia oleracea* 'Auroch', Arugula *Eruca sativa* 'Astro', and Mizuna Brassica *Brassica rapa var. japonica* production was feasible using GREENBOX technology. We processed the data collected and used descriptive statistics to demonstrate our results.

3. Results

By the 30th day of the growth cycle using GREENBOX technology for fresh crop production, we found that it was able to provide the required environmental conditions for the successful production of *Brassica rapa var. chinensis* 'Li Ren Choi', Spinach *Spinacia oleracea* 'Auroch', Arugula *Eruca sativa* 'Astro', and Mizuna Brassica *Brassica rapa var. japonica*. We determined the DLI ranged from 7.3 to 9.9 mol/m²·d with an average of 8.3 mol/m²·d which fell within the

minimum recommended range of 6.5 - 9.7 mol/m²·d [41]. The temperature varied between 20.7 °C and 27.2 °C with an average value of 24.1 °C. The VPD in each GREENBOX unit varied between 0.37 and 1.99 kPa with an average value of 1.4 kPa. The relative humidity over the growth period varied between 38.6% and 88.0% with an average value of 54.0%.

We compared our collected biomass data with biomass data from previous studies that grew the same crop species to determine the technical feasibility of crops grown using GREENBOX technology. Table 1 summarizes the average wet weights, dry weights, total chlorophyll contents, total leaf counts, and specific leaf areas (SLA).

"Li Ren Choi" crops grown using GREENBOX technology had the second largest wet weight output at 292.79 g. A study conducted by Song *et al.* [42] grew "Li Ren Choi" crops in soil for 45 days which yielded crops with an average wet weight of 100 - 110 g. Utilizing GREENBOX technology, we grew "Li Ren Choi" crops that were 187.79 grams heavier than crops grown using soil in relatively the same growth period. Regarding "Li Ren Choi", this difference in biomass indicated that GREENBOX technology is a comparable if not superior method cultivation for this crop with respect to wet weight.

With a wet weight of 194.21 g, "Auroch" Spinach produced the smallest wet weight value of all four crops using GREENBOX technology. Although "Auroch" Spinach had the smallest wet weight when evaluating the biomass performance of all four crops, another study by Janeczko *et al.* [43] grew "Auroch" Spinach with a wet weight of 151.2 g. The study conducted by Janeczko *et al.* [43] also utilized a hydroponic nutrient delivery system, but produced crops that were 43.01 g less than crops grown utilizing GREENBOX technology.

At harvest, "Astro" Arugula produced crops with a wet weight of 197.71 g. Under similar conditions, a study by Silva *et al.* [44] grew "Astro" Arugula crops to full bloom in soil. After 37 days of growth using traditional soil-based cultivation, the study recorded crops with an average wet weight of 37.2 g. Utilizing GREENBOX technology provided ideal environmental conditions and nutrient delivery which resulted in "Astro" Arugula crops 160.51 g heavier than the crops grown using the traditional method of soil. GREENBOX technology was able to grow crops that met or exceeded the biomass performance of crops grown in other fashions in less time.

The wet weights of each crop varied from 194.21 to 1031.71 g, with Mizuna Brassica (*Brassica rapa var. Japonica*) producing the highest wet weight. A similar study that grew Mizuna Brassica produced a crop with a wet weight of 162.58 g [45]. Utilizing GREENBOX technology, we were able to grow Mizuna Brassica to a wet weight 869.13 g heavier than in the study conducted by Adlloğlu *et al.* [45]. Another study that grew Mizuna Brassica (*Brassica rapa var. Japonica*) in soil for 45 days, produced an average wet weight value of 49.66 g [46]. Utilizing GREENBOX technology for the cultivation of Mizuna Brassica produced a wet weight value that was 982.05 g heavier than the soil grown Mizuna Brassica in

relatively the same growth period. A superior wet weight indicates that Mizuna Brassica can be viably grown using GREENBOX technology to a biomass that exceeds average wet weights of Mizuna Brassica grown in other fashions.

When evaluating the biomass performance of the four chosen crops over the complete growth period, we found that each crop had a consistent growth rate. We observed no indication of inhibited growth factors such as disease and pests at any point in the 30-day growth period. **Figure 2** illustrates the wet and dry weight growth trends of the crops over the 30-day growth period to produce our chosen crops using GREENBOX technology.



Figure 2. Wet and dry weight (g) trends of *Brassica rapa var. chinensis* 'Li Ren Choi', Spinach *Spinacia oleracea* 'Auroch', Arugula *Eruca sativa* 'Astro' and Mizuna Brassica *Brassica rapa var. japonica* using GREENBOX technology over the course of the 30-day growth period. **Table 1.** Wet weight (g), dry weight (g), total leaf count (n), specific leaf area (SLA) (cm²/g), and total chlorophyll content (mg/cm²) of the four crops grown using GREENBOX technology.

Plant Species	Wet Weight	Dry Weight	Leaf Count	Specific Leaf Area (SLA)	Total Chlorophyll Content
	(g)	(g)	(n)	(cm ² /g)	(mg/cm ²)
Li Ren Choi <i>"Brassica rapa var. Chinensis</i> "	292.79	12.23	33	219.67	0.055
Auroch Spinach " <i>Spinacia oleracea</i> "	194.21	11.28	50	248.13	0.043
Mizuna Brassica " <i>Brassica rapa var. japonica</i> "	1031.71	58.54	167	269.05	0.037
Astro Arugula " <i>Eruca sativa</i> "	197.71	14.89	60	202.48	0.051

4. Conclusion

The main aim of this study was to determine the technical feasibility and performance of different leafy vegetable crop production using GREENBOX technology. Our results have determined that GREENBOX technology may provide optimal environmental conditions and successfully produce "Li Ren Choi", "Auroch" Spinach, "Astro" Arugula, and Mizuna Brassica. All four crops grew to full bloom with superior, if not comparable, biomass productivity in the 30-day growth period. Since all four crops grew to full bloom and reached a superior wet weight value, we can conclude that GREENBOX technology is a viable cultivation method for these leafy green vegetable crop species. Further studies may investigate the production of fruiting or ornamental crops using GREENBOX technology.

Acknowledgements

We would like to thank Christal Niemeyer for her assistance in coordinating and acquiring the supplies necessary to complete this project, and Dr. Greg Tolley of The Water School at FGCU for providing a space to perform this project and financial assistance. We also thank Dr. Minh Nguyen and the Honors College, Dr. Heather Skaza-Acosta and the Whitaker Center.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

 Pison, G. (2017) Tous les pays du monde (2017). *Population & Sociétés*, 547, 1-8. <u>https://doi.org/10.3917/popsoc.547.0001</u>

- Maisonet-Guzman, O.E. (2011) Food Security and Population Growth in the 21st Century. <u>https://www.e-ir.info/2011/07/16/arefailing-and-failed-states-a-post-cold-war-phenomenon/</u>
- [3] Specht, K., Siebert, R., Hartmann, I., Freisinger, U.B., Sawicka, M., Werner, A., Dierich, A., *et al.* (2014) Urban Agriculture of the Future: An Overview of Sustainability Aspects of Food Production in and on Buildings. *Agriculture and Human Values*, **31**, 33-51. <u>https://doi.org/10.1007/s10460-013-9448-4</u>
- [4] Taghizadeh, R. (2021) Assessing the Potential of Hydroponic Farming to Reduce Food Imports: The Case of Lettuce Production in Sweden. Master's Thesis, Uppsala universitet, Uppsala.
- [5] Ben Hassen, T. and El Bilali, H. (2022) Impacts of the Russia-Ukraine War on Global Food Security: Towards More Sustainable and Resilient Food Systems? *Foods*, 11, Article 2301. <u>https://doi.org/10.3390/foods11152301</u>
- [6] Organisation for Economic Co-Operation and Development (2022) Economic and Social Impacts and Policy Implications of the War in Ukraine. OECD Economic Outlook, Interim Report, March 2022. <u>https://www.oecd-ilibrary.org/sites/4181d61b-en/index.html?itemId=/content/publication/4181d61b-en</u>
- [7] Hendrickson, D., Smith, C. and Eikenberry, N. (2006) Fruit and Vegetable Access in Four Low-Income Food Deserts Communities in Minnesota. *Agriculture and Human Values*, 23, 371-383. <u>https://doi.org/10.1007/s10460-006-9002-8</u>
- [8] Hamelin, A.M., Beaudry, M. and Habicht, J.P. (2002) Characterization of Household Food Insecurity in Québec: Food and Feelings. *Social Science and Medicine*, 54, 119-132. <u>https://doi.org/10.1016/S0277-9536(01)00013-2</u>
- Olson, C.M. (1999) Symposium: Advances in Measuring Food Insecurity and Hunger in the U.S. Introduction. *The Journal of Nutrition*, **129**, 521S-524S. <u>https://doi.org/10.1093/in/129.2.521S</u>
- [10] Zenk, S.N., Schulz, A.J., Israel, B.A., James, S.A., Bao, S. and Wilson, M.L. (2005) Neighborhood Racial Composition, Neighborhood Poverty, and the Spatial Accessibility of Supermarkets in Metropolitan Detroit. *American Journal of Public Health*, 95, 660-667. <u>https://doi.org/10.2105/AJPH.2004.042150</u>
- [11] Raja, S., Ma, C. and Yadav, P. (2008) Beyond Food Deserts: Measuring and Mapping Racial Disparities in Neighborhood Food Environments. *Journal of Planning Education and Research*, 27, 469-482. <u>https://doi.org/10.1177/0739456X08317461</u>
- [12] Sugrue, T.J. (2014) The Origins of the Urban Crisis. Princeton University Press, Princeton.
- [13] Ball, K., Timperio, A. and Crawford, D. (2009) Neighbourhood Socioeconomic Inequalities in Food Access and Affordability. *Health and Place*, **15**, 578-585. <u>https://doi.org/10.1016/j.healthplace.2008.09.010</u>
- [14] Chung, C. and Myers, S.L. (1999) Do the Poor Pay More for Food? An Analysis of Grocery Store Availability and Food Price Disparities. *Journal of Consumer Affairs*, 33, 276-296. <u>https://doi.org/10.1111/j.1745-6606.1999.tb00071.x</u>
- Yang, W., Dall, T.M., Beronjia, K., Lin, J., Semilla, A.P., Chakrabarti, R., Petersen, M.P., *et al.* (2018) Economic Costs of Diabetes in the U.S. in 2017. *Diabetes Care*, 41, 917-928. <u>https://doi.org/10.2337/dci18-0007</u>
- [16] Hammond, R. and Levine (2010) The Economic Impact of Obesity in the United States. *Diabetes, Metabolic Syndrome and Obesity: Targets and Therapy*, 2010,

285-295. https://doi.org/10.2147/DMSOTT.S7384

- [17] Oldani, C. (2020) The Multiple Benefits of Urban Agriculture: Contexts and Contributions of a Modern Food Movement. *Vanderbilt Undergraduate Research Journal*, **11**, 86-102. <u>https://doi.org/10.15695/vurj.v11i1.5059</u>
- [18] Kloas, W., Groß, R., Baganz, D., Graupner, J., Monsees, H., Schmidt, U., Rennert, B., et al. (2015) A New Concept for Aquaponic Systems to Improve Sustainability, Increase Productivity, and Reduce Environmental Impacts. Aquaculture Environment Interactions, 7, 179-192. https://doi.org/10.3354/aei00146
- [19] Conforti, P., Alexandratos, N., Anriquez, G., Baffes, J., Beintema, N., Boedeker, G. and Bruinsma, J. (2011) Looking Ahead in World Food and Agriculture: Perspectives to 2050. World Food and Agriculture to 2030/2050 Revisited. Highlights and Views Four Years Later. <u>http://www.fao.org/docrep/014/i2280e/i2280e.pdf</u>
- [20] Kozai, T. (2018) Current Status of Plant Factories with Artificial Lighting (PFALs) and Smart PFALs. In: Kozai, T., Ed., *Smart Plant Factory: The Next Generation Indoor Vertical Farms*, Springer, Singapore, 3-13. https://doi.org/10.1007/978-981-13-1065-2_1
- [21] Hodges, C.N., Groh, J.E. and Johnson, A.W. (1968) Controlled-Environment Agriculture for Coastal Desert Areas. *Proceedings of National Agricultural Plastics Conference*, 8, 58-68.
- [22] Gómez, C., Currey, C.J., Dickson, R.W., Kim, H.J., Hernández, R., Sabeh, N.C., Burnett, S.E., *et al.* (2019) Controlled Environment Food Production for Urban Agriculture. *HortScience*, **54**, 1448-1458. <u>https://doi.org/10.21273/HORTSCI14073-19</u>
- [23] Benke, K. and Tomkins, B. (2017) Future Food-Production Systems: Vertical Farming and Controlled-Environment Agriculture. *Sustainability: Science, Practice, and Policy*, 13, 13-26. <u>https://doi.org/10.1080/15487733.2017.1394054</u>
- [24] Kozai, T. (2013) Resource Use Efficiency of Closed Plant Production System with Artificial Light: Concept, Estimation and Application to Plant Factory. *Proceedings* of the Japan Academy, Series B, 89, 447-461. <u>https://doi.org/10.2183/pjab.89.447</u>
- [25] Goodman, W. and Minner, J. (2019) Will the Urban Agricultural Revolution Be Vertical and Soilless? A Case Study of Controlled Environment Agriculture in New York City. *Land Use Policy*, 83, 160-173. <u>https://doi.org/10.1016/j.landusepol.2018.12.038</u>
- [26] Yang, X., Theobald, D., McAvoy, R., Wu, J. and Liu, C. (2017) Greenbox Farming: A New System for Urban Agriculture. 2017 ASABE Annual International Meeting, Spokane, 16-19 July 2017, 1 p.
- [27] Singh, A.K. and Yang, X. (2021) GREENBOX Horticulture, an Alternative Avenue of Urban Food Production. *Agricultural Sciences*, **12**, 1473-1489. <u>https://doi.org/10.4236/as.2021.1212094</u>
- [28] Singh, A.K., McAvoy, R.J., Bravo-Ureta, B. and Yang, X. (2021) An Experimental Study on GREENBOX Technology: Feasibility and Performance. 2021 ASABE Annual International Virtual Meeting, 12-16 July 2021, 145-166. https://doi.org/10.13031/aim.202100453
- [29] Singh, A.K., McAvoy, R., Bravo-Ureta, B. and Yang, X. (2023) GREENBOX Technology I—Technical Feasibility and Performance in Warehouse Environment. *Journal of the ASABE.*
- [30] Singh, A.K., McAvoy, R.J., Bravo-Ureta, B. and Yang, X. (2021) Comparison of Environmental Condition, Productivity, and Resources Use between GREENBOX and

Greenhouse for Growing Lettuce. 2021 ASABE Annual International Virtual Meeting, 12-16 July 2021, 2-10. <u>https://doi.org/10.13031/aim.202100455</u>

- [31] Singh, A.K., Bravo-Ureta, B., McAvoy, R. and Yang, X. (2023) GREENBOX Technology II—Comparison of Environmental Conditions, Productivity, and Water Consumption with Greenhouse Operation. *Journal of the ASABE.*
- [32] Singh, A.K., Bravo-Ureta, B. and Yang, X. (2022) Financial Feasibility Study of GREENBOX Technology for Crop Production in an Urban Setting. 2022 ASABE Annual International Meeting, Houston, 17-20 July 2022, 1-16. https://doi.org/10.13031/aim.202201068
- [33] Duever, M.J., Meeder, J.F., Meeder, L.C. and McCollom, J.M. (1994) The Climate of South Florida and Its Role in Shaping the Everglades Ecosystem. In: Davis, S. and Ogden, J.C., Eds., *Everglades: The Ecosystem and Its Restoration*, CRC Press, Boca Raton, 225-248.
- [34] National Oceanic and Atmospheric Administration (2022) Climate—Southwest Florida. <u>https://www.weather.gov/wrh/Climate?wfo=tbw</u>
- [35] Fitz-Rodríguez, E., Kubota, C., Giacomelli, G.A., Tignor, M.E., Wilson, S.B. and McMahon, M. (2010) Dynamic Modeling and Simulation of Greenhouse Environments under Several Scenarios: A Web-Based Application. *Computers and Electronics in Agriculture*, **70**, 105-116. <u>https://doi.org/10.1016/j.compag.2009.09.010</u>
- [36] Kozai, T, Niu, G. and Takagaki, M. (2015) Plant Factory: An Indoor Vertical Farming System for Efficient Quality Food Production. Academic Press, Cambridge. <u>https://books.google.com/books?id=z-C7DwAAQBAJ</u>
- [37] Liu, H., Fu, Y., Wang, M. and Liu, H. (2017) Green Light Enhances Growth, Photosynthetic Pigments and CO₂ Assimilation Efficiency of Lettuce as Revealed by "Knock Out" of the 480-560 nm Spectral Waveband. *Photosynthetica*, 55, 144-152. https://doi.org/10.1007/s11099-016-0233-7
- [38] Zhu, J., Tremblay, T. and Liang, Y. (2012) Comparing SPAD and atLEAF Values for Chlorophyll Assessment in Crop Species. *Canadian Journal of Soil Science*, **92**, 645-648. <u>https://doi.org/10.4141/cjss2011-100</u>
- [39] Anderson, C.J.R. and Rosas-Anderson, PJ. (2017) Leafscan (Version 1.3.21). https://itunes.apple.com/app/id1254892230
- [40] Richardson, A.D., Duigan, S.P. and Berlyn, G.P. (2002) An Evaluation of Noninvasive Methods to Estimate Foliar Chlorophyll Content. *New Phytologist*, 153, 185-194. <u>https://doi.org/10.1046/j.0028-646X.2001.00289.x</u>
- [41] Paz, M., Fisher, P.R. and Gómez, C. (2019) Minimum Light Requirements for Indoor Gardening of Lettuce. Urban Agriculture & Regional Food Systems, 4, 1-10. <u>https://doi.org/10.2134/urbanag2019.03.0001</u>
- [42] Song, L., Luo, H., Jiang, L., Hou, J., Zhang, T., Dai, L. and Yu, Z. (2020) Integrative Analysis of Transcriptome and Metabolome Reveals the Possible Mechanism of Leaf Yellowing in Pak Choi (*Brassica rapa* subsp. *chinensis*) with 1-Methylcyclopropene Treatment during Storage at 20°C. *Postharvest Biology and Technology*, 169, Article ID: 111300. <u>https://doi.org/10.1016/j.postharvbio.2020.111300</u>
- [43] Janeczko, D.B. and Timmons, M.B. (2019) Effects of Seeding Pattern and Cultivar on Productivity of Baby Spinach (*Spinacia oleracea*) Grown Hydroponically in Deep-Water Culture. *Horticulturae*, 5, Article 20. https://doi.org/10.3390/horticulturae5010020
- [44] Silva, P.A.D., Kinjo, S., Melo, M.P.B.X.D. and Sala, F.C. (2019) Evaluation of Arugula Cultivars and Seed Production in the Organic System. *Journal of Seed Science*,

41, 423-430. https://doi.org/10.1590/2317-1545v41n4218457

- [45] Adİloğlu, S., Açıkgöz, F.E. and Adİloğlu, A. (2015) The Effect of Increasing Doses of N Application on Some Agronomic Characteristics, Vitamin C, Protein and Mineral Content of Mibuna (*Brassica rapa* var. *Nipposinica*) and Mizuna (*Brassica rapa* var. *Japonica*) Plants. *Ziraat Fakültesi Dergisi, Uludağ Üniversitesi*, 29, 1-11.
- [46] Hasturk Sahin, F., Aktas, T., Eryilmaz Acikgoz, F. and Akcay, T. (2016) Some Technical and Mechanical Properties of Mibuna (*Brassica rapa* var. *Nipposinica*) and Mizuna (*Brassica rapa* var. *Japonica*). *PeerJ PrePrints*, 4, e1698v1. https://doi.org/10.7287/peerj.preprints.1698v1