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Arugula and Lettuce Responses to Greenhouse Hydroponic Systems: An Analysis of Yield and Resource Use Efficiencies

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Abstract. **Controlled environment agriculture (CEA) for leafy green production is growing in popularity as consumers and producers become more environmentally conscious. Growers can use several hydroponic systems in CEA leafy green production, but detailed information about their tradeoffs is scarce. In this experiment, we sought to grow leafy greens in several soilless systems to evaluate yield and resource use and determine optimal production systems. Arugula (*Eruca sativa* 'Astro') and lettuce (*Lactuca sativa* 'Casey') were grown in a greenhouse in four soilless systems: deep water culture (DWC), nutrient film technique (NFT), vertical tower, and aeroponics. System resource inputs in terms of water, energy, and area, as well as system plant outputs like biomass production and nutrient concentrations were quantified during the 28-day experiment. Water (WUE), energy (EUE), and area use efficiencies (AUE) were then calculated using fresh shoot biomass. Based on these resource use efficiencies (WUE, EUE, and AUE), arugula had excellent performance in aeroponics, satisfactory performance in both the NFT and vertical systems, and comparatively poor performance in DWC. Lettuce performed satisfactorily in both DWC and NFT, but both systems had reduced EUE, and DWC also had reduced AUE. In contrast, lettuce had fair performance in the vertical and aeroponic systems. In conclusion, arugula is optimally grown in aeroponics, and lettuce is optimally grown in DWC or NFT. Understanding these system tradeoffs will help growers and the CEA industry to become more sustainable and profitable.**

Leafy greens are consumed daily by millions of people around the world. Some of the most popular leafy green crops globally are lettuce (*Lactuca sativa*), spinach (*Spinacia oleracea*), and cabbage and relatives such as cauliflower and brussels sprouts (all *Brassica oleracea*). China produced the most lettuce in 2022, with 14.98 million metric tons, while the United States was the second highest producer, with 3.30 million metric tons (Food and Agriculture Organization 2024). California (73%) and Arizona (21%) account for nearly all lettuce produced in the United States each year (Davis et al. 2023). The combined lettuce production from these two states in 2022 was valued at about $4.12 billion (US Department of Agriculture 2023).

The leafy green industry in the United States has faced significant challenges in recent years, attributed to a confluence of factors such as drought, heat, and pest pressures. Data from 2018 to 2022 indicates that field lettuce yields, particularly in California, have been lower than historical averages (Davis et al. 2023). Furthermore, extreme weather in the spring of 2023 led to delayed planting and flooded fields in the Salinas Valley, a region responsible for 60% of US lettuce production. The agricultural commissioner's office in Monterey County (where the Salinas Valley is located) stated that this extreme weather caused an estimated $324.1 million in crop damage (Lee 2023). Extreme weather events around the globe are projected to both intensify and become more frequent in the coming decades due to climate change; field agriculture as we know it is at risk (Bolster et al. 2023). Due to these climate challenges and the increasing cost of labor and other production inputs, interest in controlled environment agriculture (CEA) has risen in recent years.

CEA, broadly, shields food production from unfavorable weather. Companies can employ CEA technology to various degrees, such as low-technology structures like low or high tunnels, medium-technology greenhouses with simple irrigation and ventilation management, high-technology greenhouses with advanced climate control, and indoor vertical farms or plant factories. CEA not only insulates crops from the adverse effects of climate change but can also decrease the need for pesticides while increasing the growing season length for areas with suboptimal climates. This lower dependency of pesticides can be further reduced by using soilless hydroponic growing systems to produce specific crops in CEA such as strawberries (*Fragaria* ×*ananassa*) (Wrenn et al. 2023). CEA growing systems can also enhance overall crop quality while maximizing yield (Gómez et al. 2019). Over 25,000 t of lettuce was grown using CEA in the United States in 2019, valued at about $71 million. For the same year, the CEA production area increased by 28%, and yield increased by more than 50% compared with 2014. Nearly 66% of this CEA lettuce was produced using hydroponic systems (Davis et al. 2023).

Hydroponic systems use nutrient solutions rather than soil to deliver essential nutrients to plant roots, and water culture systems are the most popular type of hydroponic system for growing leafy greens (Gómez et al. 2019). The crop roots primarily interact with a liquid nutrient solution instead of solid soil or soilless substrate in water culture systems. This solution can be in constant contact with the roots (statically or dynamically), or the roots can be intermittently exposed to the solution via flowing or misting. These systems provide several advantages compared with soilless substrate systems and traditional field production for leafy greens beyond the advantage of being insulated from extreme weather events. A previous study reported that lettuce grown in water culture systems showed significantly improved yield, nutrition, and water use efficiency compared with lettuce grown in a soil-based system (Majid et al. 2021). Meaningful improvements in lettuce fresh biomass and nutrient content in two water culture systems compared with lettuce grown in a sand substrate culture system have also been reported (El-Helaly and Darwish 2019). Furthermore, it was found that two water culture systems outperformed a substrate culture system when examining lettuce fresh shoot biomass (Li et al. 2018). It seems clear that water culture systems are likely the optimal cultivation method for lettuce production in CEA.

Lettuce is a popular hydroponic crop with high consumer demand due to its multiple uses in the food industry. Arugula (*Eruca sativa*) is another popular leafy green previously cultivated in water culture hydroponic systems (Bonasia et al. 2017; Houston et al. 2023; Mainos et al. 2023; Yang et al. 2021). Lettuce and arugula are common additions to mixed salad products and accumulate several secondary metabolites, such as glucosinolates and vitamin C, that are beneficial to human health (Costa-Pérez et al. 2023; Hall et al. 2012; Jilani et al. 2015).

With the rising interest in CEA globally, it is more important than ever that growers select the production system that can maximize both their fiscal and environmental sustainability. There are several types of hydroponic water culture systems, with the primary difference among them being the nature of the interaction between the nutrient solution and the plant roots. The simplest system is a deep water culture (DWC) system (also known as a floating raft culture system) wherein the plants are suspended above a static pool of nutrient solution with aeration. In a nutrient film technique (NFT) system, the plants are grown in a sloped channel. The nutrient solution is pumped into the elevated end of the channel and then flows down the channel through the plant roots, creating a constant film of nutrient solution along the bottom of the channel. An aeroponic system uses a high-pressure pump and nozzles to mist the nutrient solution onto the roots of the plant. Both NFT and aeroponic systems are recirculating systems, and these two systems, along with DWC, are the most used water culture systems (Gómez et al. 2019). There is a fourth type of system that has grown in popularity in recent years with producers: the vertical system. This system stacks plants vertically in a tower that can be hollow or filled with a wicking substrate. The nutrient solution is pumped to the top of the tower, where it can then percolate downward and interact with the plant roots.

Several studies have examined the differences in yield and/or resource use between two water culture systems for lettuce production in CEA (El-Helaly and Darwish 2019; El-Shinawy et al. 1996; El-Ssawy et al. 2020; Gillani et al. 2023; Lennard and Leonard 2006; Li et al. 2018). However, a comprehensive comparison of DWC, NFT, aeroponic, and vertical systems considering plant performance (biomass/yield and quality) and several resource use efficiencies (water, energy, and footprint area) has not been undertaken. Furthermore, there is a lack of literature concerning any water culture system comparisons for arugula production, let alone one that is comprehensive as previously described.

The objective of this study was to execute just such a comprehensive system comparison for both lettuce and arugula in water culture hydroponic systems. This includes comparing the absolute outputs, such as fresh and dry biomass, and the ratio of outputs to inputs, i.e., water, energy, and footprint area, for each system. These ratios are the resource use efficiencies. By understanding the tradeoffs in both absolute and relative outputs among these systems, producers can make informed decisions about growing systems as the leafy green CEA industry matures.

Material and Methods

Location and environmental conditions

This experiment was conducted at the University of Georgia (College of Agricultural and Environmental Sciences, Department of Horticulture, Controlled Environment Agriculture Crop Physiology and Production Laboratory) in Athens, GA, USA (lat. 33°55′55.10″N, long. 83°21′50.51″W, altitude 198 m) from Jun to Jul 2023 in a polycarbonate greenhouse with temperature control using a pad-fan cooling system and unit heaters for heating.

Greenhouse air temperature and relative humidity were monitored using a digital sensor (HMP60; Vaisala, Helsinki, Finland) connected to a datalogger (CR1000X; Campbell Scientific, Logan, UT, USA) for automatic data collection and had average ± standard error values of 25.7 ± 0.19 °C and 75.9 ± 1.01%, respectively. Vapor pressure deficit (VPD) was calculated using this temperature and relative humidity data and was 0.92 ± 0.047 kPa. Canopy-level light was measured by a quantum sensor (SQ-610; Campbell Scientific) connected to a separate datalogger (CR1000; Campbell Scientific) and resulted in a daily light integral (DLI) of 20.2 ± 1.16 $mol \cdot m^{-2} \cdot d^{-1}$. The plants were subject only to natural sunlight during the experiment duration.

Plant material

Pelleted seeds of lettuce ‘Casey’ and nonpelleted seeds of arugula ‘Astro’ were purchased from a commercial seed supplier (Johnny’s Selected Seeds, Winslow, ME, USA) and sown in 2.5-× 2.5-× 4-cm rockwool blocks (AO 25/40; Grodan, Roermond, The Netherlands) with one lettuce seed per block and four to eight arugula seeds per block. Sown seeds were placed in a walk-in growth chamber with 23.5 ± 0.01 °C temperature, 58.3 ± 0.65% relative humidity, 1.2 ± 0.02 kPa VPD, 14.4 $mol \cdot m^{-2} \cdot d^{-1}$ DLI, and 847 ± 11.2 $mg \cdot L^{-1}$ CO_2 for 14 d under daily automated ebb and flow subirrigation to allow for germination and initial growth. After 14 d, similarly sized plants were randomly selected to be transplanted into one of the four hydroponic systems. Arugula was thinned to four seedlings per rockwool block at transplanting.

Hydroponic system

For the experiment, lettuce and arugula were grown in four different water culture hydroponic systems: DWC, NFT, vertical tower, and aeroponic (Fig. 1). Each system was considered a treatment, and the DWC system was the control, all with four replications. The number of plants per system varied, but the same number within each treatment and replication was measured and analyzed. Four plants for both crops were randomly selected from each system for measurement and analysis.

Each experimental unit of the DWC system had 69 plants for each crop in 2.699 m^2 for a density of 25.57 plants/m^2, the NFT system had 72 plants for each crop in 1.6722 m^2 for a density of 43.06 plants/m^2, the vertical system had 36 plants for each crop in 0.5602 m^2 for a density of 64.26 plants/m^2, and the aeroponic system had 42 plants for each crop in

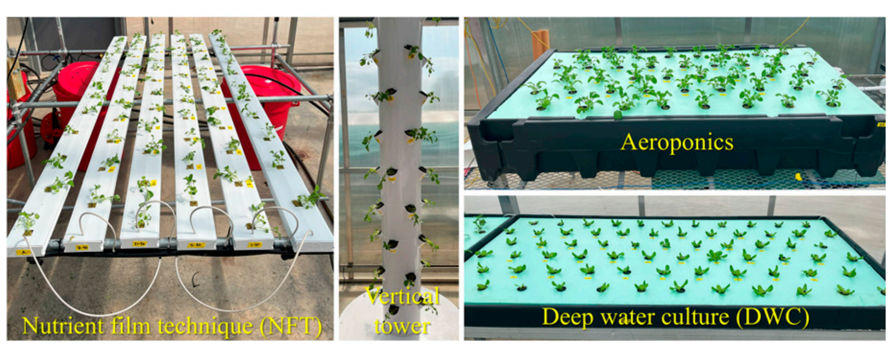

Fig. 1. Photos showing the soilless systems tested in this experiment: deep water culture (DWC), nutrient film technique (NFT), vertical tower, and aeroponics.

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2.09 m^2 for a density of 20.10 plants/m^2. The three horizontal systems (DWC, NFT, and aeroponic) had 20.3-cm plant spacing. The DWC, NFT, and vertical systems had separate reservoirs for each crop, while the aeroponic system had a shared reservoir due to the pressure needed to operate the nozzles. Nonedge sites in each system were numbered as potential measurement sites.

DWC system. The DWC system was constructed from commercially available products using two 242.5-× 121-× 17-cm grow trays (OD; Botanicare, Vancouver, WA, USA) to serve as the reservoirs and foam insulation boards (GreenGuard LG 0.75"; Kingspan, Kingscourt, Ireland) cut to size to make floating rafts with drilled holes for net cups measuring 4.8 cm tall with 4.5-cm top diameters and 3.3-cm bottom diameters (Teku G46; Pöppelmann GmbH & Co., Lohne, Germany). The solution in the grow trays was aerated using air stones connected to a 13.5 $m^3 \cdot h^{-1}$, 48 kPa, 1.27-cm outlet aeration pump (EcoAir 7; EcoPlus, Vancouver, WA, USA), with six stones per tray. Edge plants surrounded trays, and each crop had one tray with 38 potential measurement sites per replication.

NFT system. The NFT system was assembled from commercially available products and consisted of 244-× 11.5-× 4-cm channels (CHA9008; Crop King, Lodi, OH, USA) with corresponding tops (CHA9004) and end caps (CHA9100 and CHA9101) from the same manufacturer as the channels. A 121-L plastic reservoir (H-3687; Uline, Pleasant Prairie, WI, USA) held the nutrient solution recirculated by a submersible pump (PE-1; Little Giant, Oklahoma City, OK, USA). Each channel was on a 1% slope and had 12 prepunched, square holes for the rockwool blocks. Each crop had six channels, and edge plants surrounded channels. The middle four channels for each crop together contained 40 potential measurement sites per replication. The two edge channels for each crop were fertigated by separate reservoirs to simplify system management. All pumps ran continuously throughout the experiment.

Vertical system. The vertical system was a commercially available hydroponics system (Tower Garden Flex; Tower Garden, Collierville, TN, USA). It consisted of a 75-L, bowl-shaped reservoir with a submersible pump (Syncra 3.0; Sicce, Pozzoleone, Italy) on the bottom and a columnar tower on top. This tower was made of nine layers, each 18 cm tall, with four plant sites around its circumference. An inner, central column connected to the reservoir pump on the bottom reached above the top layer to the stop cap. As the pump activated, nutrient solution was sent up this column, hit the stop cap, and then percolated down through every layer into the reservoir by gravity. The area between the central column and the outer wall, where the sites for the plants were located, was hollow, allowing roots to interact with the nutrient solution as it percolated. There were also holes just below the stop cap and at every layer that aided with even distribution as the solution percolated. Each crop had one tower of 36 plants per replication. The plant sites in each tower's top and bottom layers were considered edge sites, giving 28 potential measurement sites for each crop. Nutrient solution delivery was controlled by a timer (TGT1; Tower Garden), which activated the pumps for 3 min on and 12 min off in alternating intervals throughout the experiment.

Aeroponic system. The aeroponic system consisted of a mix of commercial and custom-built equipment: two black plastic tubs sloped on the inside and with grooves in the lips to hold cut-to-size foam insulation board tops (GreenGuard LG 0.75"; Kingspan) with drilled holes for net cups (Teku G46; Pöppelmann GmbH & Co.). Manifolds for the inside of each tub were constructed out of PVC piping and 16 misting nozzles (22219221202; Tefen, Kibbutz Nahsholim, Israel), which were supplied with a nutrient solution by high-pressure pump (EF1000; Everflo Pumps, Paynesville, MN, USA). Nutrient solution delivery was controlled by the same timer as the vertical system and was on the same interval regime. Edge plants surrounded trays, and each crop had 18 potential measurement sites per replication.

Fertilization

A modified Sonneveld solution was used for fertigation in all systems (Sonneveld and Kreij 1987). The solution contained (all values in $mg \cdot L^{-1}$): 152 total N with 140 NO_3-N and 12 NH_4-N, 31 P, 221 K, 90 Ca, 26 Mg, 32 S, 0.25 B, 0.5 Cu, 1.8 Fe, 0.13 Mn, 0.02 Mo, and 0.16 Zn.

Measurements

Reservoir pH, electrical conductivity (EC), temperature, and dissolved oxygen (DO). The solution pH and EC were measured regularly with a digital probe (#HI98131; Hanna Instruments, Smithfield, RI, USA) and adjusted to maintain between 5.5 and 6.5 pH and between 1.25 and 1.75 $dS \cdot m^{-1}$, respectively. A commercial product derived from phosphoric acid was used to reduce the pH of the solution (pH Down; Advanced Nutrients, West Hollywood, CA, USA), while an 8 M solution of potassium hydroxide was used to raise the solution pH. EC was lowered by diluting the solution with tap water. Reservoir DO was measured using a digital probe (HI98193; Hanna Instruments) at 4 and 13 d after transplanting (DATs).

Nondestructive plant measurements. Leaf chlorophyll content (expressed as chlorophyll content index) and leaf anthocyanin content (expressed as anthocyanin content index) were measured using handheld meters (CCM-200 plus and ACM-200 plus, respectively; Opti-Sciences, Hudson, NH, USA) on one of the youngest, fully mature leaves per plant at 16 DATs. Arugula plant height was measured using a meter stick at 17 DATs. Chlorophyll content, anthocyanin content, and height were measured for all plants in each replication. One plant per replication was used to measure the gas exchange parameters stomatal conductance, carbon assimilation, evapotranspiration, and water use efficiency (WUE_G) using a portable photosynthesis system (CIRAS 4; PP Systems, Amesbury, MA, USA). Arugula gas exchange parameters were measured at 20 DATs, and lettuce gas exchange parameters were measured at 25 DATs. Analysis dates were decided based on plant morphology and physiology coupled with our availability to process the samples in a single day with the team available.

Destructive plant measurements. Arugula was harvested at 21 DATs, and lettuce at 28 DATs based on plant morphology and maturation when plants were considered salable. Fresh biomass was measured for every plant in each replication using digital scales for the fresh shoot biomass (#PB3002; Mettler Toledo, Griefensee, Switzerland) and the fresh root biomass (item 30430061; Ohaus Corporation, Parsippany, NJ, USA). Fresh shoot biomass was used in subsequent calculations to indicate yield. One plant in each replication for both crops was used to measure total soluble solids (TSSs) by selecting two to three leaves and crushing them in a garlic press with cheesecloth onto a digital refractometer (#HI96801; Hanna Instruments). Three plants in each replication for both crops were deconstructed and scanned using a leaf area meter (LI-3100; LI-COR, Lincoln, NE, USA) to obtain the total leaf area. For two arugula plants per replication and three lettuce plants per replication, shoots and roots were placed into paper bags and dried in an oven at 80 °C until completely dry. The dried tissue was weighed on the same digital scales used to obtain the dry biomass of shoots and roots. Shoot water content was calculated using the fresh and dry biomass measurements, and the shoot-to-root ratio (shoot:root) was calculated by dividing shoot dry biomass by root dry biomass. One plant per replication for both crops was placed into a sample bag after being weighed for fresh biomass and sent to a commercial laboratory (Waters Agricultural Laboratories, Camilla, GA, USA) for tissue nutrient concentration analysis. Leaf N was determined by high temperature combustion process (Nelson and Sommers 1973). Leaf P, K, Ca, Mg, S, B, Cu, Fe, Mn, and Zn concentrations were determined by inductively coupled plasma atomic emission spectrophotometer after wet acid digestion using nitric acid and hydrogen peroxide (Twyman 2005).

System measurements and resource use quantification. T-type thermocouples (T/C wire 20 gauge; Antylia Scientific, Vernon Hills, IL, USA) were used to measure reservoir temperature every hour using the same datalogger as the quantum sensor for automatic data collection. The reservoir volumes were tracked throughout the experiment in all systems. All reservoirs were filled to a known volume at transplanting and filled again to that known volume after draining and refilling. Residual reservoir volume was measured during drain and refill events, which were triggered when reservoir volume was low and/or when the pH and EC were extremely out of the ideal ranges (5.5 to 6.5 for pH and 1.25 to 1.75 $dS \cdot m^{-1}$ for EC). Volume was measured using a 25-mm digital flow meter (O-WM-2; Restmo, Staten Island, NY, USA). By knowing reservoir volume before and after refills, total system losses due to evapotranspiration (ET) were easily calculated by simple subtraction.

To calculate plant water use efficiency (WUE), ET per plant was first calculated by dividing reservoir ET by the number of plants supplied by that reservoir. Plant fresh shoot biomass was then divided by this ET per plant (based on which system the plant was harvested) to obtain plant WUE (in $g \cdot L^{-1}$). Note that the DWC, NFT, and vertical systems had separate reservoirs for each crop, while the aeroponic system had a shared reservoir due to the pressure needed to operate the nozzles.

Total system energy use was calculated by tracking the total pump activation time in hours for each system. The NFT recirculating pump and the DWC aerating pump ran continuously, whereas the vertical and aeroponic pumps ran on the 3 min on and 12 min off cycle, which is a 20% activation rate. Power consumption (in watts) of all pumps was determined based on manufacturer specifications (200 W for DWC, 36 W for NFT, 48 W for vertical, and 48 W for aeroponic). The pump run time and power consumption were multiplied to obtain total system energy use in kilowatt hours (kWh). Note that the DWC aeration pump and aeroponic fertigation pump were shared between the two crops.

To calculate plant energy use efficiency (EUE), energy use per plant was first calculated by dividing the total system energy use by the number of plants in that system. Fresh shoot biomass was then divided by this energy use per plant to obtain plant EUE (in $g \cdot kWh^{-1}$).

The system footprint area was calculated by measuring the widest system dimensions with a tape measure and calculating the footprint area appropriately (the footprint areas of the DWC, NFT, and aeroponic systems are rectangular, and the vertical system is circular). One DWC tray (69 plants total), one single NFT channel (12 plants), one vertical tower (36 plants), and one aeroponic tub (42 plants) were measured for these calculations.

System area use efficiency (AUE), or the maximum yield per area that a particular system can deliver, was calculated by multiplying fresh shoot weight by the number of plants per measured system (or system component as outlined in the previous paragraph) and then dividing that resulting number by the system footprint area. Note that the AUE calculation does not consider the spacing between systems needed to implement these systems at scale effectively but instead represents an ideal maximum AUE.

Experimental design and statistical analysis

We tested four hydroponics systems (DWC, NFT, vertical, and aeroponic) with four replications arranged on a randomized block design, with treatments assigned as fixed effects. Statistical analysis was performed by conducting one-way analysis of variance (ANOVA) with Tukey's post-hoc test using statistical software (SigmaPlot version 15; Systat Software, San Jose, CA, USA) to determine significant differences among treatments. When a data set did not meet the ANOVA's normality or equal variance distribution conditions, a Kruskal–Wallis test with Dunn's post-hoc was conducted using the same statistical software. A significance level of 0.05 or 5% was used in all tests. Results from each crop were analyzed separately.

Results

Reservoir pH, EC, temperature, and DO. The two crops had different effects on reservoir pH over the growing cycle. Arugula caused reservoir pH in all four systems to increase consistently, becoming more alkaline and above the ideal range of 5.5 to 6.5 throughout the experiment (Fig. 2A). This trend became particularly strong from 15 to 20 DATs. On the contrary, lettuce caused variations in alkaline and acidic directions for all systems except for the aeroponic system (Fig. 2B). The NFT system for lettuce had extreme variations in both directions. The average measured reservoir pH for arugula was 6.6 ± 0.10, 6.6 ± 0.13, 6.6 ± 0.10, and 6.8 ± 0.16 in the DWC, NFT, vertical, and aeroponic systems, respectively, and for lettuce it was 6.1 ± 0.15, 5.8 ± 0.19, 6.1 ± 0.13, and 6.6 ± 0.15 in the same systems. Conversely, the two crops had similar effects on reservoir EC for all systems (Fig. 2C and 2D). All systems start at the bottom of the ideal range and then trend upwards through around 7 DATs. After 7 DATs, there is a drop in EC through 11 DATs, with the tower vertical system diverging from the three horizontal systems. A substantial decrease in EC occurred for both crops in the aeroponic system at 20 DATs, with the arugula NFT also reaching a minimum at this point. After lettuce EC in all systems trends slightly downward from 11 DATs through 25 DATs, there is a sharp increase followed immediately by a sharp decrease in all systems in the final 4 days before harvest. The average ± standard error measured reservoir EC for arugula was 1.3 ± 0.03, 1.3 ± 0.05, 1.4 ± 0.04, and 1.2 ± 0.07 $dS \cdot m^{-1}$, and for lettuce it was 1.3 ± 0.03, 1.3 ± 0.03, 1.4 ± 0.04, and 1.2 ± 0.06 $dS \cdot m^{-1}$ for the DWC, NFT, vertical, and aeroponic systems, respectively, for both crops.

Average hourly reservoir temperature was significantly affected by the systems for both crops. For arugula, the DWC and NFT systems had a 4.2% higher average reservoir temperature than both the aeroponic and vertical systems, and the aeroponic system had a 2.6% higher average reservoir temperature than the vertical system ($P < 0.001$) (Fig. 3A). For lettuce, each system had a significantly different average hourly temperature; the DWC had the highest average reservoir temperature at 28.8 °C, the NFT temperature was second highest at 28 °C, the aeroponic temperature was third highest at 26.7 °C, and the vertical temperature was the lowest at 26.6 °C ($P < 0.001$) (Fig. 3B). Reservoir DO was consistent for both crops in all systems, with all measurements falling between 5.5 and 7.75 $mg \cdot L^{-1}$ on both days (Fig. 3C and 3D).

Leaf pigment indices. For arugula, the NFT, vertical, and aeroponic systems reduced leaf chlorophyll ($P < 0.001$) (Fig. 4A) and anthocyanin content ($P < 0.001$) (Fig. 4C) by 43% and 34%, respectively, compared with the DWC control. There were no significant differences in either pigment index among those three recirculating systems. Lettuce showed no significant differences in chlorophyll content for any system ($P = 0.364$) (Fig. 4B), while anthocyanin content was 9.1% higher in both the aeroponic and NFT systems compared with the vertical system ($P = 0.001$) (Fig. 4D).

Height and gas exchange parameters. Arugula plant height was 24% higher in the three recirculating systems (NFT, vertical, and aeroponic) compared with the DWC control, and there were no significant differences among these three recirculating systems ($P < 0.001$) (Fig. 5). Arugula carbon assimilation was 75% higher in the aeroponic system compared with the vertical system ($P = 0.037$) (Table 1). Arugula stomatal conductance ($P = 0.586$), evapotranspiration ($P = 0.383$), and WUE_G ($P = 0.235$) were not significantly affected by the treatments. Furthermore, for lettuce, neither stomatal conductance ($P = 0.881$), carbon assimilation ($P = 0.170$), evapotranspiration ($P = 0.693$), nor WUE_G ($P = 0.554$) were affected by the treatments (Table 1).

Fresh biomass. Arugula fresh shoot biomass was 48% higher in the three recirculating systems compared with the DWC control ($P < 0.001$), and there were no significant differences among the three recirculating systems (Fig. 6A). The aeroponics system showed 50% higher fresh root biomass than the vertical system, and there were no other significant differences for arugula fresh root biomass ($P = 0.029$) (Fig. 6C). Lettuce fresh shoot biomass was 30% higher in the DWC control compared with the vertical system ($P < 0.001$) (Fig. 6B). There were no other significant differences for fresh shoot biomass. There were also no significant differences among the systems for lettuce fresh root biomass ($P = 0.420$) (Fig. 6D).

TSS and leaf area. There were no significant differences in TSS for either arugula ($P = 0.067$) (Fig. 7A) or lettuce among any of the systems ($P = 0.663$) (Fig. 7B). For arugula, the three recirculating systems resulted in 58% higher leaf area compared with the DWC system ($P < 0.001$); there were no significant differences in leaf area among these three systems (Fig. 7C). The lettuce leaf area from the aeroponic system was 13% lower than each of the three other systems (DWC, NFT, and vertical) ($P < 0.001$) (Fig. 7D).

Dry biomass, shoot water content, and shoot:root. Arugula dry shoot biomass was 61% higher in the aeroponic and NFT systems than in the DWC control ($P = 0.002$) (Fig. 8A). Conversely, lettuce dry shoot biomass was 13% higher in the DWC system

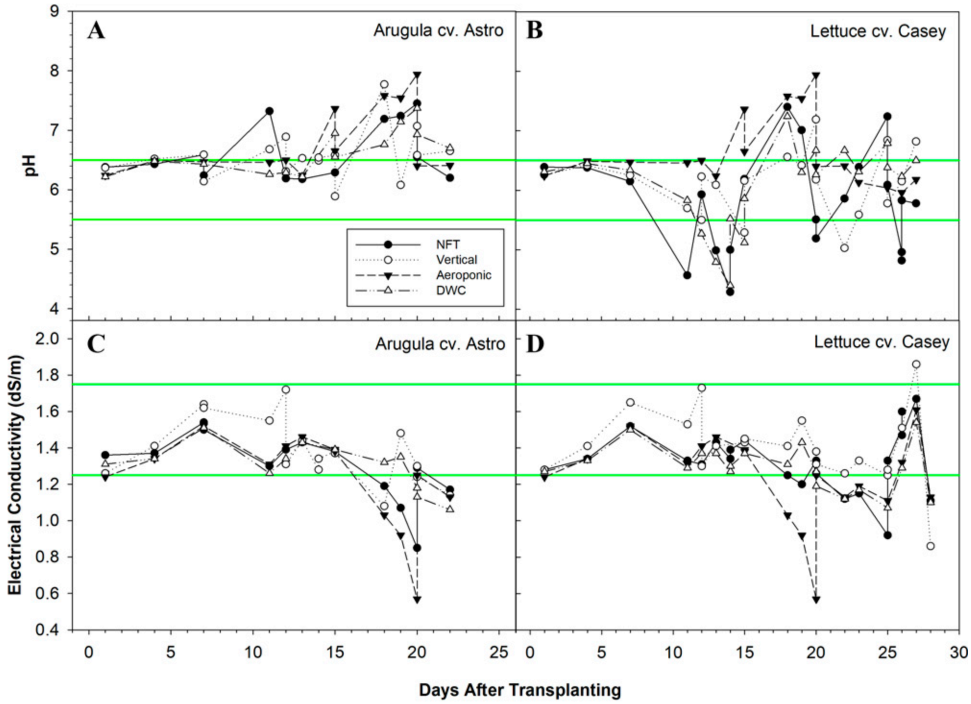

Fig. 2. Arugula (*E. sativa* 'Astro') reservoir pH (**A**), lettuce (*L. sativa* 'Casey') reservoir pH (**B**), arugula reservoir electrical conductivity (EC) (**C**), and lettuce reservoir EC (**D**) from 0 to 21 d after transplant (DATs) for arugula and 0 to 28 DATs for lettuce. Individual data points represent pH or EC measurements at that time point. Green lines represent the bounds of the ideal range (5.5 to 6.5 for pH and 1.25 to 1.75 for EC); data points between the green lines are considered within range. DWC = deep water culture; NFT = nutrient film technique.

than in the aeroponic and vertical systems ($P = 0.003$) (Fig. 8B). Arugula dry root biomass was 91% higher in the aeroponics system than in the DWC control and NFT systems ($P = 0.003$) (Fig. 8C). Lettuce dry root biomass was 22% higher in the vertical system compared with the DWC system ($P = 0.016$) (Fig. 8D). Arugula shoot water content was 1.4% lower in the DWC system than in both the NFT and vertical systems ($P < 0.001$) (Fig. 8E). Lettuce shoot water content was 0.45% lower in the vertical system than in each of the three horizontal systems ($P < 0.001$) (Fig. 8F). Arugula shoot:root was 117% higher in the NFT system than in the aeroponic system ($P = 0.021$) (Fig. 8G). Lettuce shoot:root was 31% higher in the DWC system than in the other three systems ($P < 0.001$) (Fig. 8H).

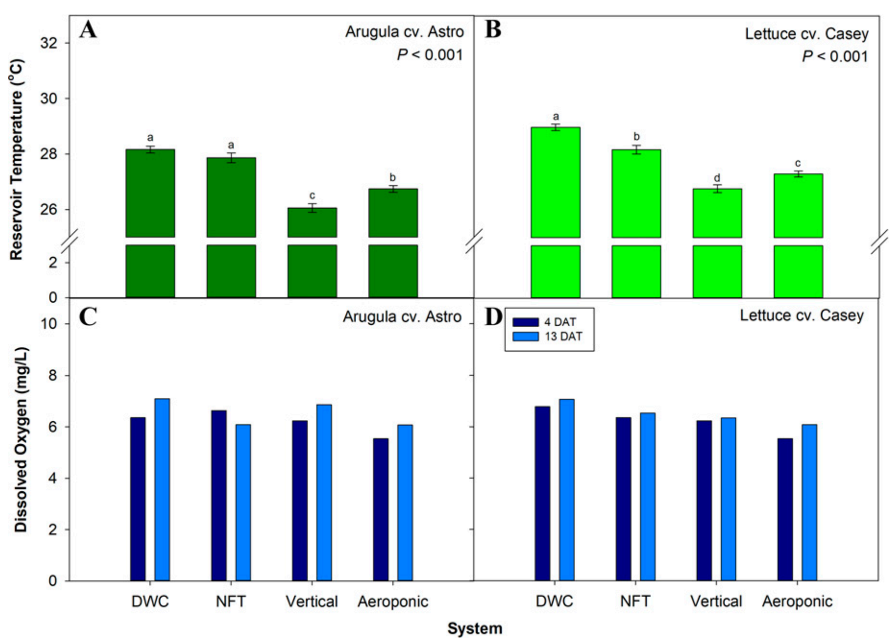

Fig. 3. Reservoir temperature for arugula (*E. sativa* 'Astro') (**A**) and lettuce (*L. sativa* 'Casey') (**B**) in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems. Each bar represents the average ± standard error of four replications with four measurement plants; bars with the same letter show no significant difference; bars with different letters show significant differences. All tests executed using significance level of 5% ($P < 0.05$). Reservoir dissolved oxygen at four and 13 d after transplanting (DATs) for arugula (**C**) and lettuce (**D**). Each bar represents one measurement.

Leaf tissue macronutrient concentration. The treatments significantly affected arugula leaf N ($P = 0.047$); however, the post-hoc test did not distinguish between treatments at the 0.05 level (Table 2). The treatments did not significantly affect arugula leaf P ($P = 0.470$). Arugula leaf K was 24% higher in the vertical and aeroponic systems when compared with the DWC control ($P = 0.001$). Arugula leaf Mg was 31% higher in the aeroponic system compared with the DWC control ($P = 0.009$). Arugula leaf Ca was not significantly affected by the systems ($P = 0.205$). Finally, arugula leaf S was 18% higher in the aeroponic system than in the three other systems ($P < 0.001$). Lettuce leaf N in the vertical system was 26% higher than in the DWC system ($P = 0.030$) (Table 2). Lettuce leaf P was 43% higher in both the vertical and aeroponic systems compared with the DWC control ($P = 0.006$). The treatments significantly affected lettuce leaf K, but the post-hoc test could not distinguish between the treatments ($P = 0.046$). Lettuce leaf Mg was 16% higher in the aeroponics system than in the three other systems ($P < 0.001$). Also, lettuce leaf Mg was 19% higher in the NFT system compared with the DWC control. The treatments

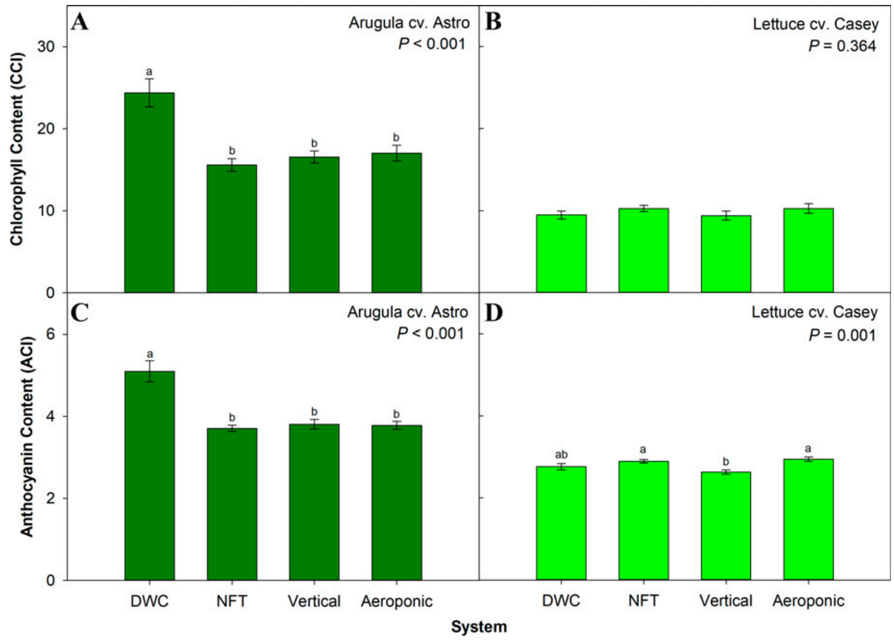

Fig. 4. Chlorophyll content for arugula (*E. sativa* 'Astro') (**A**) and lettuce (*L. sativa* 'Casey') (**B**), and anthocyanin content for arugula (**C**) and lettuce (**D**) in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems at 16 d after transplant. Each bar represents the average ± standard error of four replications with four measurement plants; bars with the same letter show no significant difference; bars with different letters are statistically different at 5% probability ($P < 0.05$). ACI = anthocyanin content index; CCI = chlorophyll content index.

did not significantly affect lettuce leaf Ca ($P = 0.111$) and leaf S ($P = 0.118$).

Leaf tissue micronutrient concentration. Arugula leaf Mn was 31% higher in both the NFT and vertical systems when compared with the aeroponic system ($P = 0.010$) (Table 3). The treatments significantly affected arugula leaf Zn ($P = 0.042$) and Cu ($P = 0.046$), but the post-hoc tests could not distinguish between treatments. The treatments did not significantly affect arugula leaf B ($P = 0.374$) and Fe ($P = 0.446$). Lettuce leaf Zn was 27% higher in both the DWC control and vertical systems when compared with the aeroponic system ($P = 0.006$) (Table 3). Lettuce leaf B ($P = 0.743$), Mn ($P = 0.114$), Fe ($P = 0.698$), and Cu ($P = 0.076$) were not significantly affected by the treatments.

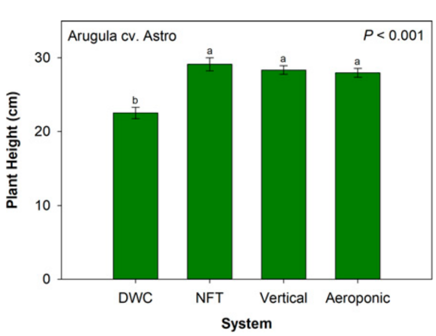

Fig. 5. Arugula (*E. sativa* 'Astro') plant height measured using a meter stick at 17 d after transplanting in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems. Each bar represents the average ± standard error of four replications with four measurement plants. Bars with the same letter show no significant difference; bars with different letters are statistically different at 5% probability ($P < 0.05$).

Resource use efficiencies. Arugula WUE was 29% higher in the aeroponic system than in all other systems (Fig. 9A). Furthermore, WUE from the NFT system was 41% higher than from both the DWC and vertical systems for arugula ($P < 0.001$). Lettuce WUE was 21% higher in the DWC and NFT systems than in the vertical or aeroponic systems ($P < 0.001$) (Fig. 9B). Both arugula and lettuce had comparable results for EUE. The aeroponic system resulted in 514% and 532% higher EUE for arugula (Fig. 9C) and lettuce (Fig. 9D), respectively, than the DWC and NFT systems. The vertical system also had 704% and 318% higher EUE for arugula and lettuce, respectively, than the DWC system ($P < 0.001$). Both crops also had similar results for AUE. The vertical and NFT systems had 197% and 52% higher AUE for arugula (Fig. 9E) and lettuce (Fig. 9F), respectively, than the DWC and aeroponic systems ($P < 0.001$).

Discussion

Both arugula and lettuce had similar responses in fresh shoot biomass production (our indicator of yield) in the three recirculating systems: the NFT system had the highest biomass out of the three, with the aeroponic system having the second most, and the vertical system having the lowest biomass of the three (Fig. 6A and 6B). The crops responded very differently to the DWC system, however. For arugula, the fresh shoot biomass from the DWC system was the lowest of any system, but for lettuce, the DWC produced the highest fresh shoot biomass of any system.

A comparative reduction in root zone oxygen availability is one possible explanation for this reduction in arugula fresh shoot biomass in the DWC system. Root respiration will decrease if roots are not supplied with oxygen at a sufficient volume and rate, harming plant growth and development (Roblero et al. 2020). In the three recirculating systems, the area around the roots is never completely occupied by nutrient solution, and there are gaps for ambient air to interact with the roots. In the DWC system, the plants are embedded in a floating foam raft in constant contact with the nutrient solution, leaving no air gaps for roots to interact with the ambient atmosphere. Although the DWC nutrient solution was aerated and DO was measured to be sufficiently high based on (Ferrarezi et al. 2024) to avoid negative impacts on root respiration at 4 and 13 DATs (Fig. 3C and 3D), the oxygen uptake rate by the plants could have exceeded this rate of oxygen input from the aeration, particularly as the plants matured and oxygen requirements increased. This theory is corroborated by a previous study by our research group in the same greenhouse growing arugula in a similar DWC system that saw a reduction in DO for arugula over a 21-d growth cycle in the summer (Ferrarezi et al. 2024). Furthermore, DO measurements in this experiment were made on the edge of the system, and there could be localized depletion zones in the system that were not detected. A higher degree of granularity in DO measurement would be required to fully assess its role in reducing

Table 1. Stomatal conductance (gs), carbon assimilation (A), evapotranspiration (E), and water use efficiency (WUE_G) of arugula (*E. sativa* 'Astro') and lettuce (*L. sativa* 'Casey') in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems at 20 d after transplanting.

Crop	System	gs (mmol H_2O m^{-2} s^{-1})	A (μmol CO_2 m^{-2} s^{-1})	E (mmol H_2O m^{-2} s^{-1})	WUE_G (μmol CO_2 $mmol^{-1}$ H_2O)
Arugula	DWC	579.40 ± 162.894	13.40 ± 1.276 ab	3.41 ± 0.636	4.01 ± 0.449
	NFT	735.68 ± 141.973	13.78 ± 1.785 ab	3.45 ± 0.218	4.08 ± 0.719
	Vertical	501.52 ± 118.860	8.47 ± 1.011 b	2.93 ± 0.366	3.04 ± 0.496
	Aeroponic	686.11 ± 73.152	14.83 ± 1.542 a	2.89 ± 0.104	5.20 ± 0.712
P		0.586	0.037	0.383	0.235
Lettuce	DWC	477.73 ± 109.797	7.83 ± 1.403	3.90 ± 0.375	1.96 ± 0.231
	NFT	518.39 ± 94.391	11.65 ± 1.144	4.50 ± 0.275	2.61 ± 0.286
	Vertical	413.38 ± 64.907	7.87 ± 1.116	4.49 ± 0.478	1.92 ± 0.542
	Aeroponic	427.61 ± 130.843	9.70 ± 1.448	4.29 ± 0.420	2.29 ± 0.353
P		0.881	0.170	0.693	0.544

Statistical analysis for gs, A, and E was conducted using one-way analysis of variance (ANOVA) and Tukey's honestly significant difference (AN), while the Kruskal–Wallis test with Dunn's post-hoc (KW) was used for WUE_G due to invalid ANOVA assumptions. All tests were executed using a significance level of 5% ($P < 0.05$). Columns with the same letter within the same crop show no significant difference.

arugula fresh shoot biomass in the DWC system.

Reservoir temperature also likely contributed to the arugula fresh shoot biomass reduction in the DWC system. Elevated root zone temperatures have been previously shown to negatively affect arugula fresh shoot biomass in hydroponic systems, even at temperatures as low as 28 °C (He et al. 2022; Lai and He 2016). In the case of the DWC system, the reservoir temperature is the same as the root zone temperature since the roots are in constant contact with the nutrient solution. The average hourly reservoir temperature for arugula in the DWC system was above 28 °C (Fig. 3A), which could be enough to induce heat stress and thus negatively affect growth. The NFT system had a similar average reservoir temperature to the DWC system for arugula, but fresh shoot biomass was not reduced similarly (Fig. 6A and 6B). This is likely because the NFT system is a recirculating system, which allows roots to cool more effectively under its forced convection conditions. The static DWC system relies on natural convection alone for root cooling, a less efficient heat transfer method than forced convection (Çengel and Ghajar 2020). Higher temperature solutions also have reduced DO solubility and increased oxygen consumption by plants from the solution (Bozorg-Haddad et al. 2021; Hendrickson et al. 2022). It is likely that the combination of reduced DO availability due to system architecture, solution temperature-induced heat stress, and a further reduction in DO availability due to the elevated solution temperature caused the decrease in arugula fresh shoot biomass in the DWC system.

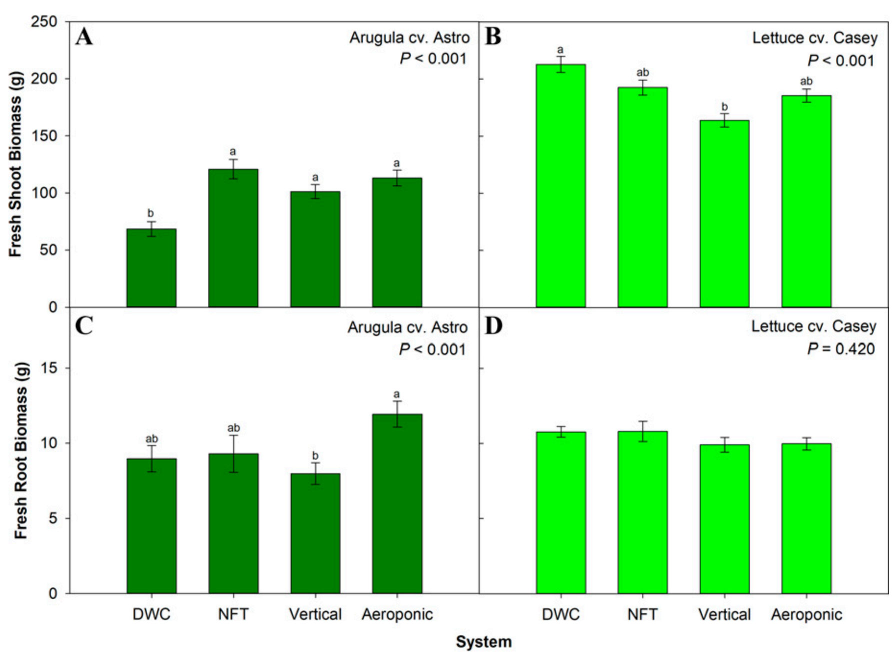

Fig. 6. Fresh shoot biomass for arugula (*E. sativa* 'Astro') (**A**) and lettuce (*L. sativa* 'Casey') (**B**), and fresh root biomass for arugula (**C**) and lettuce (**D**) in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems at 21 and 28 d after transplant, respectively, for arugula and lettuce. Each bar represents the average ± standard of four replications with four measurement plants. Bars with the same letter show no significant difference; bars with different letters are statistically different at 5% probability ($P < 0.05$).

Our results for lettuce fresh shoot biomass are consistent with previously published studies. 'Green Oak' lettuce has been reported to produce greater fresh shoot biomass in a DWC system than an NFT system for a 21-DAT harvest (Lennard and Leonard 2006). We also saw improved lettuce fresh shoot biomass production in our DWC system compared with our NFT system (Fig. 3B). Other studies have reported greater fresh shoot biomass from NFT systems than from aeroponic systems for various lettuce cultivars and experiment lengths from transplant to harvest (El-Shinawy et al. 1996; El-Ssawy et al. 2020; Li et al. 2018). Our results differ in that lettuce fresh shoot biomass production was similar between the NFT and aeroponic systems. This result resembles a study that reported no statistical difference in yield between aeroponic and NFT systems for 'Red fire' lettuce for a 42-DAT harvest (El-Helaly and Darwish 2019). The variation in results for aeroponic system fresh shoot biomass production is likely due to a large variation in aeroponic system design, with many factors that contribute to plant performance, such as droplet size, total volumetric flow rate, volumetric flow rate per plant, and frequency of spraying. Research into how these factors influence lettuce growth in aeroponic systems has recently been investigated, but results have been varied and inconsistent (Lakhiar et al. 2019, 2018; Tunio et al. 2022).

Fresh root biomass for arugula was the highest in the aeroponic system (Fig. 3C), which matches results from several other crops that showed enhanced root growth in aeroponic systems (Lakhiar et al. 2018). A likely contributing factor to this increase in root growth is greater access to root zone oxygen since the roots were hanging in the air for 12 min of every 15 min throughout the experiment. We also see an improvement in dry root biomass for arugula in the aeroponic system (Fig. 8C), which demonstrates that this increase in fresh root biomass was truly due to an increase in growth and not due to favorable osmotic gradients for water uptake and storage in the roots.

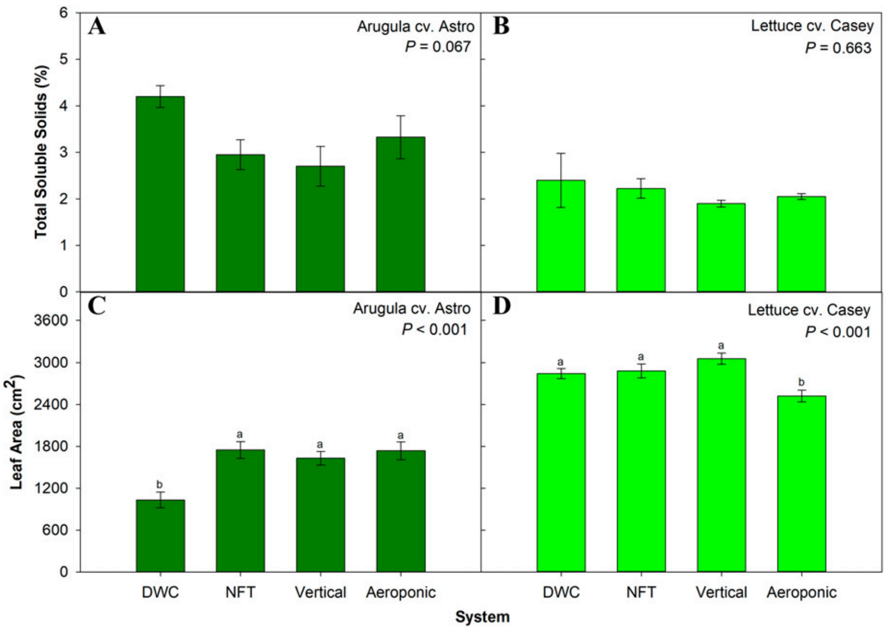

Fig. 7. Total soluble solids for arugula (*E. sativa* 'Astro') (**A**) and lettuce (*L. sativa* 'Casey') (**B**), and leaf area for arugula (**C**) and lettuce (**D**) in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems at 21 and 28 d after transplant, respectively, for arugula and lettuce. Each bar represents the average ± standard of four replications with four measurement plants. Bars with the same letter show no significant difference; bars with different letters are statistically different at 5% probability ($P < 0.05$).

The systems did not significantly affect lettuce fresh root biomass (Fig. 6D), which is not an unexpected result. Previous studies have reported higher fresh root biomass from aeroponic systems than from NFT systems; however, the differences were not statistically significant (El-Helaly and Darwish 2019; El-Ssawy et al. 2020; Li et al. 2018). Of those three studies, only one reported significantly higher lettuce dry root biomass from the aeroponic system, which does not match our result (Li et al. 2018).

Foliar tissue nutrient concentrations varied according to the growing system: for both crops, N, K, and Mg were significantly affected, with the lowest values for all three nutrients coming from the DWC system (Table 3). Both crops also had the lowest P concentration in the DWC system, although only the result from lettuce was significant. The fact that both crops had similar foliar analysis results makes it likely that static hydroponic systems affect nutrient uptake in leafy greens. One possible explanation for these results is that in static hydroponic systems, the solution immediately around plant roots could deplete nutrients, creating a boundary layer-like dead zone that can only be resupplied by diffusion. This phenomenon would worsen as the crop matures due to increased uptake, root growth, and nutrient competition. One possible consequence of this localized nutrient depletion in the DWC system is a reduction in the ability of the plants to regulate cell ψ_S and turgidity. Solutes generally, and potassium in particular, are vital for cell osmoregulation. By regulating the influx and efflux of solutes across membranes and between tissues, plants can manipulate water potentials and control water flow across membranes and between tissues (Taiz and Zeiger 2010). We observed decreased fresh shoot biomass (Fig. 6A), decreased foliar potassium concentration, and decreased shoot water content (Fig. 8E) of arugula in the DWC system, indicating that those plants could have been less able to osmoregulate and maintain turgidity due to decreased solute uptake. Increasing turgidity is one of the primary mechanisms that drives cell wall and leaf expansion in plants (Boyer 1988), so an inability to properly regulate turgidity would inhibit cell wall expansion and negatively affect growth. In lettuce, however, there was a reduction in foliar solute concentrations (Table 2) but no reduction in either fresh shoot biomass (Fig. 6B) or shoot water content (Fig. 8F); this indicates that the decrease in solute uptake in the DWC system was not detrimental to lettuce growth. This could be due to the DWC system being a more favorable environment for lettuce cell osmoregulation, which is supported by our observation of lettuce generally having a higher shoot water content than arugula. We also observed a significant increase in lettuce shoot:root in the DWC system (Fig. 7H), and lettuce shoot:root has previously been reported to decrease significantly in response to osmotic stress in hydroponic growing conditions (Moncada et al. 2020). This inverse relationship between shoot:root and osmotic stress further supports our assertion that the DWC system promotes optimal osmoregulation for lettuce.

Another interesting result from the foliar tissue analysis is that there was a significantly higher concentration of sulfur in the aeroponic system than in any other system (Table 2). Many glucosinolate compounds contain S, and glucosinolate concentrations in arugula leaves have been previously shown to vary based on solution EC in NFT systems (Costa-Pérez et al. 2023; Yang et al. 2021). Furthermore, genes and transcripts associated with glucosinolate biosynthesis have been previously shown to up-regulate in response to osmotic stress for several other species in the *Brassicaceae* family (Eom et al. 2018; Podda et al. 2019). We also observed the lowest arugula shoot:root in the aeroponic system (Fig. 8G). Arugula, like lettuce, has been seen to decrease shoot:root in response to osmotic stress in hydroponic systems (Campos et al. 2018). Our observation of low shoot:root in the aeroponic system could indicate that those plants were experiencing mild osmotic stress. It is possible that this increased S concentration in the aeroponic system could correspond to an increase in glucosinolate concentrations in response to osmotic stress; however, glucosinolate quantification would be necessary to confirm this assertion.

The differences in arugula WUE were driven by yield and average ET per plant. The vertical system had the highest average plant ET (0.986 L), the NFT system had the second highest (0.835 L), the DWC system had the third highest (0.716 L), and the aeroponic system had the lowest (0.607 L). Despite having the lowest ET per plant, the DWC system still had the lowest WUE (Fig. 9A) due to its lower fresh shoot biomass production (Fig. 6A). The vertical system had reasonably high fresh shoot biomass production, but its WUE was comparatively low due to its high ET per plant. The NFT system had the highest fresh shoot biomass, which helped to offset its second highest plant ET to result in the second highest WUE. The aeroponic system had both the lowest ET per plant and the second-highest fresh shoot biomass production, which resulted in the highest WUE of any system by a considerable margin.

Lettuce WUE (Fig. 9B) was driven primarily by fresh shoot biomass (Fig. 6B), unlike arugula WUE. This is because the magnitude of differences in average plant ET in each system was much smaller for lettuce; the aeroponic system had the highest (1.095 L), the vertical system had the second highest (0.972 L), the NFT system had the third highest (0.940 L), and the DWC system had the lowest ET per plant (0.880 L). One potential source of error in these lettuce ET calculations is that aeroponic system evaporation likely increased in the final week of growth. The arugula was harvested from the system at 21 DATs, and the nozzles continued to spray into an empty chamber until the lettuce was harvested at 28 DATs, and the system was deactivated. The empty chamber was covered during that week; however, evaporation could still have increased. One previous study reported that WUE in their NFT and aeroponic systems

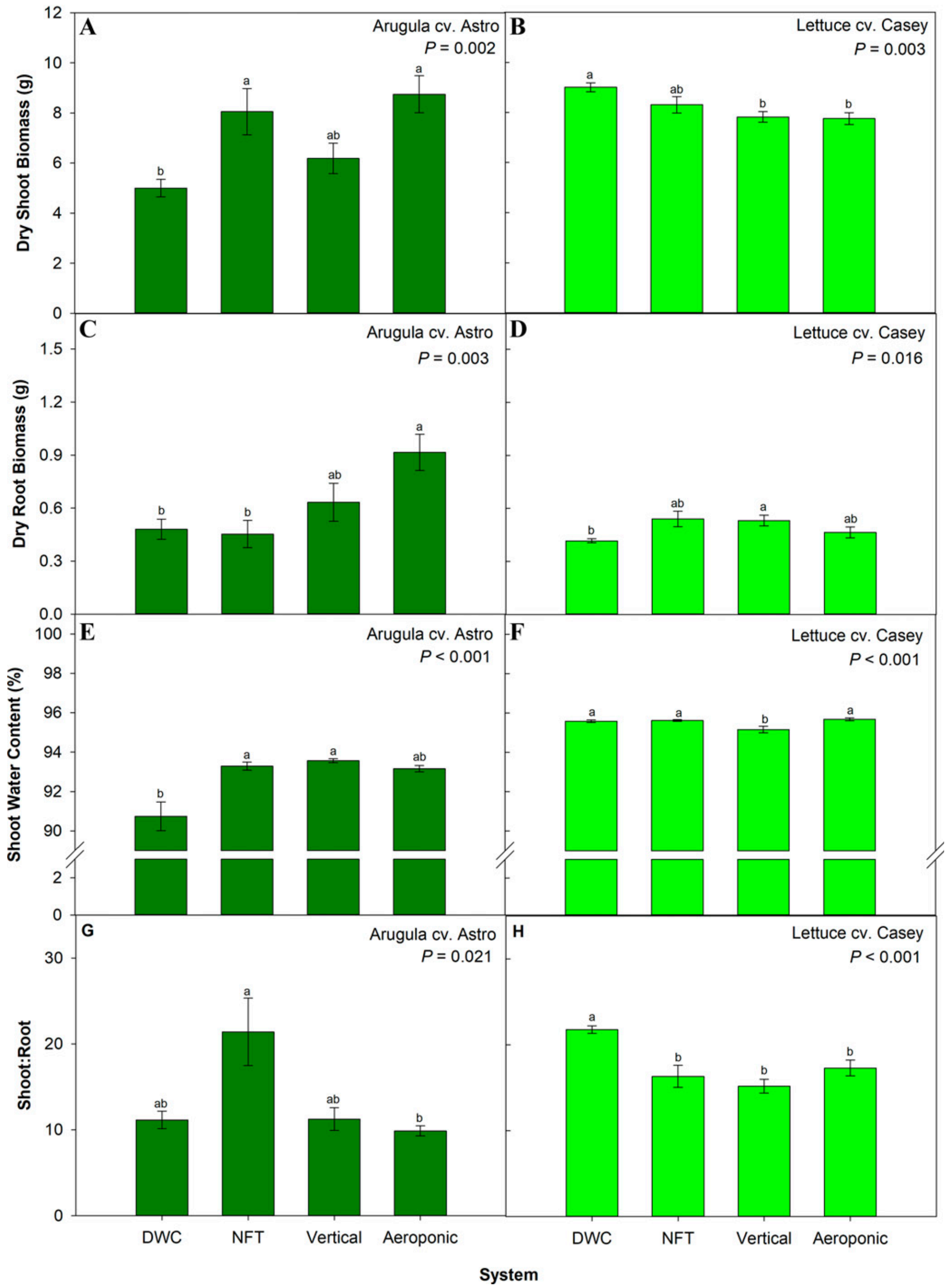

Fig. 8. Dry shoot biomass for arugula (*E. sativa* 'Astro') (**A**) and lettuce (*L. sativa* 'Casey') (**B**), dry root biomass for arugula (**C**) and lettuce (**D**), shoot water content for arugula (**E**) and lettuce (**F**), and shoot-to-root ratio (shoot:root) for arugula (**G**) and lettuce (**H**) in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems at 21 and 28 d after transplant, respectively, for arugula and lettuce. Each bar represents the average ± standard of four replications with four measurement plants. Bars with the same letter show no significant difference; bars with different letters are statistically different at 5% probability ($P < 0.05$).

were 75.50 and 75.68 $g{\cdot}L^{-1}$ for 'Paris Island' lettuce, and 69.37 and 73.39 $g{\cdot}L^{-1}$ for 'Maikonig' lettuce (El-Shinawy et al. 1996). Another study reported that in 2018, 'Limor-Hyb.' lettuce grown in aeroponic systems resulted in significantly higher WUE than lettuce grown in an NFT system; however, the next year there was not a significant difference in WUE (El-Ssawy et al. 2020). The same study reported WUE values of 50 to 90 $g{\cdot}L^{-1}$ in all treatments. Our systems resulted in larger WUE values than both studies, with values between 150 and 250 $g{\cdot}L^{-1}$ in all four systems.

EUE results for both crops (Fig. 9C and 9D) were driven almost entirely by average plant energy use instead of fresh shoot biomass. For both arugula and lettuce, the highest average plant energy use came in the DWC system (0.730 and 0.974 kWh), the second highest came in the NFT system (0.378 and 0.504 kWh), the third highest came in the vertical system (0.134 and 0.179 kWh), and the lowest came in the aeroponic system (0.058 and 0.077 kWh). Both the DWC and NFT systems had pumps running continuously, resulting in lower EUE values for both systems. The DWC system had a large (200 W) pump aerating both systems, which is why this system's energy consumption was so high, and the EUE was low for both crops. The aeroponic system had the highest EUE because there was a single 48-W pump that fertigated 84 plants total (measurement and nonmeasurement, both crops), whereas each 48-W vertical system pump fertigated only 36 plants total.

Table 2. Leaf macronutrient concentrations of arugula (*E. sativa* 'Astro') and lettuce (*L. sativa* 'Casey') in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems. Leaf nitrogen (N) was determined by high temperature combustion process. Leaf phosphorous (P), potassium (K), calcium (Ca), magnesium (Mg), and sulfur (S) were determined by inductively coupled plasma atomic emission spectrophotometer after wet acid digestion using nitric acid and hydrogen peroxide.

Crop	System	N (%)	P (%)	K (%)	Ca (%)	Mg (%)	S (%)
Arugula	DWC	5.52 ± 0.247 a	0.68 ± 0.047	5.54 ± 0.155 b	3.20 ± 0.083	0.52 ± 0.010 b	1.56 ± 0.070 b
	NFT	6.63 ± 0.061 a	0.77 ± 0.032	6.27 ± 0.215 ab	3.48 ± 0.267	0.58 ± 0.029 ab	1.61 ± 0.033 b
	Vertical	6.71 ± 0.164 a	0.77 ± 0.036	6.86 ± 0.160 a	2.88 ± 0.217	0.59 ± 0.021 ab	1.72 ± 0.057 b
	Aeroponic	6.40 ± 0.142 a	0.74 ± 0.052	6.98 ± 0.132 a	3.37 ± 0.056	0.68 ± 0.027 a	2.03 ± 0.033 a
P		0.047	0.470	0.001	0.205	0.009	<0.001
Test		KW	AN	AN	AN	AN	AN
Lettuce	DWC	5.00 ± 0.359 b	0.76 ± 0.113 b	7.22 ± 1.298 a	1.49 ± 0.097	0.42 ± 0.023 c	0.29 ± 0.009
	NFT	6.29 ± 0.086 ab	1.02 ± 0.013 ab	9.72 ± 0.186 a	1.60 ± 0.013	0.50 ± 0.006 b	0.29 ± 0.003
	Vertical	6.29 ± 0.058 a	1.09 ± 0.012 a	9.84 ± 0.163 a	1.38 ± 0.072	0.46 ± 0.017 bc	0.28 ± 0.001
	Aeroponic	6.24 ± 0.058 ab	1.10 ± 0.013 a	9.07 ± 0.210 a	1.39 ± 0.054	0.58 ± 0.017 a	0.30 ± 0.003
P		0.030	0.006	0.046	0.111	<0.001	0.118
Test		KW	KW	KW	AN	AN	KW

Statistical analysis for leaf P, K, Mg, Ca, and S was conducted using one-way analysis of variance (ANOVA) and Tukey's honestly significant difference (AN), while the Kruskal–Wallis test with Dunn's post-hoc (KW) was used for leaf Zn due to invalid ANOVA assumptions. All tests executed using significance level of 5% ($P < 0.05$). Columns with the same letter within the same crop show no significant difference.

This difference in plant-to-pump ratios led to the difference in EUE between the vertical and aeroponic systems. In commercial, large-scale systems, average plant energy use and, thus, EUE results would likely be different from those we observed in this study.

AUE results were also driven primarily by the planting density inherent to the systems, but fresh shoot biomass also played a minor role. The aeroponic system had the lowest density (20.10 plants/m^2), the DWC system had the second lowest density (25.57 plants/m^2), the NFT system had the second highest density (43.06 plants/m^2), and the vertical system had the highest planting density (64.26 plants/m^2). For lettuce, the AUE results follow this trend exactly (Fig. 9F), even though there were differences in fresh shoot biomass among the systems (Fig. 6B). For arugula, the reduction in DWC fresh shoot biomass (Fig. 6A) caused the AUE from this system to be as low as the aeroponic AUE (Fig. 9E), even though it had a higher planting density. Overall, our lettuce AUE results are higher than results obtained by a previous study that reported AUE values of 4.47 and 4.13 kg·m^{-2} for 'Green Oak' lettuce grown in DWC and NFT systems, respectively (Lennard and Leonard 2006). Our results are also higher than those reported by another study that grew 'Red fire' lettuce and produced 2.46 and 2.26 kg·m^{-2} in NFT and aeroponic systems, respectively (El-Helaly and Darwish 2019). Many factors likely contributed to these differences in results, namely growing conditions, fertilizer solution composition, length of growth cycle, cultivar used, and system measurement and design. It is also difficult to apply these AUE results to commercial-scale systems since planting density in all systems will likely change as the goal becomes production rather than research.

Conclusions

The two crops responded quite differently to the four different hydroponic systems. Arugula produced the least fresh shoot biomass in the DWC system, while lettuce produced the most fresh shoot biomass in the DWC system. For both crops, the DWC system resulted in lower concentrations of foliar nutrients like N, K, and Mg. Arugula also had elevated S concentrations in the aeroponic system, indicating a possible increase in the accumulation of beneficial phytochemicals, such as glucosinolates. WUE was influenced by yield and system design, while EUE and AUE were primarily influenced by system design. The aeroponic system performed the best for arugula production, with the only drawback being reduced AUE. The DWC and NFT systems performed well for lettuce production, with the DWC system having superior yield and WUE but also the lowest EUE and poor AUE. The NFT system had a more balanced performance, with good yield, WUE, and AUE but reduced EUE. The vertical system had the best AUE for both crops, which could be a larger advantage to growers in high-cost areas than improved WUE or EUE. Understanding these tradeoffs among different hydroponic systems will enable growers to make informed decisions about their growing systems, increasing the sustainability of CEA leafy green production.

Table 3. Leaf micronutrient concentrations of arugula (*E. sativa* 'Astro') and lettuce (*L. sativa* 'Casey') in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems. Leaf boron (B), iron (Fe), copper (Cu), manganese (Mn), and zinc (Zn) concentrations were determined by inductively coupled plasma atomic emission spectrophotometer after wet acid digestion using nitric acid and hydrogen peroxide.

Crop	System	B (mg/kg)	Cu (mg/kg)	Mn (mg/kg)	Fe (mg/kg)	Zn (mg/kg)
Arugula	DWC	60.33 ± 0.882	11.67 ± 0.333 a	71.33 ± 2.906 ab	107.00 ± 8.888	143.00 ± 9.292 a
	NFT	62.00 ± 1.354	17.75 ± 3.772 a	83.50 ± 3.175 a	106.50 ± 7.100	138.75 ± 15.085 a
	Vertical	56.75 ± 3.326	14.25 ± 0.629 a	81.25 ± 5.121 a	108.25 ± 7.052	100.25 ± 12.311 a
	Aeroponic	60.67 ± 1.453	13.00 ± 0.577 a	61.00 ± 3.215 b	93.00 ± 0.577	95.67 ± 6.009 a
P		0.347	0.046	0.010	0.446	0.042
Test		AN	AN	AN	AN	AN
Lettuce	DWC	35.50 ± 6.198	15.00 ± 2.380	127.00 ± 48.030	243.25 ± 85.565	86.50 ± 4.735 a
	NFT	29.00 ± 0.408	14.25 ± 0.250	61.25 ± 3.146	121.50 ± 9.456	73.25 ± 1.109 ab
	Vertical	27.75 ± 0.750	15.00 ± 0.408	64.00 ± 5.115	107.25 ± 1.887	88.25 ± 4.589 a
	Aeroponic	29.25 ± 1.109	18.00 ± 0.480	44.25 ± 4.644	110.50 ± 5.679	68.00 ± 3.629 b
P		0.743	0.076	0.114	0.698	0.006
Test		KW	KW	KW	KW	AN

Statistical analysis was conducted using one-way analysis of variance and Tukey's honestly significant difference (AN), while the Kruskal–Wallis test with Dunn's post-hoc (KW) was used for leaf Zn due to invalid ANOVA assumptions. All tests executed using significance level of 5% ($P < 0.05$). Columns with the same letter within the same crop show no significant difference.

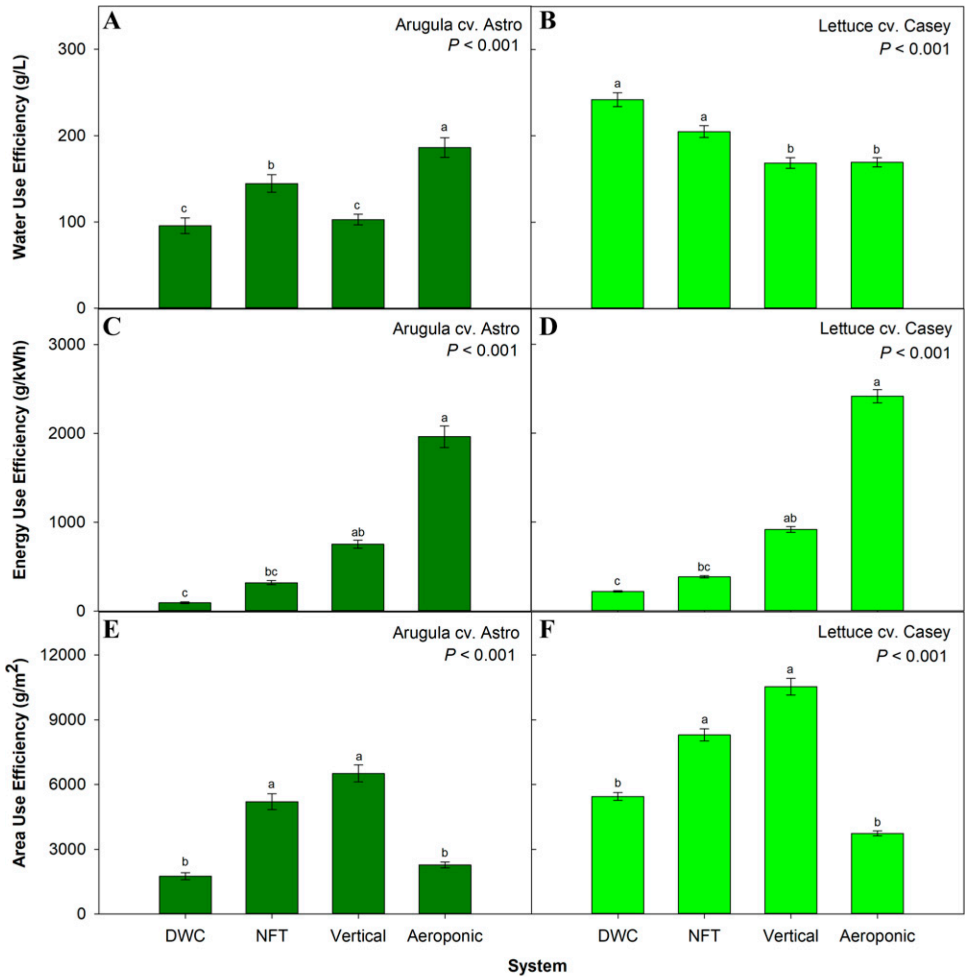

Fig. 9. Water use efficiency for arugula (*E. sativa* 'Astro') (**A**) and lettuce (*L. sativa* 'Casey') (**B**), energy use efficiency for arugula (**C**) and lettuce (**D**), and area use efficiency or the maximum yield per area that a particular system can deliver for arugula (**E**) and lettuce (**F**) in the deep water culture (DWC), nutrient film technique (NFT), vertical, and aeroponic systems at 21 and 28 d after transplant, respectively, for arugula and lettuce. Each bar represents the average ± standard of four replications with four measurement plants. Bars with the same letter show no significant difference; bars with different letters are statistically different at 5% probability ($P < 0.05$).

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