

# Effects of Two Planting Densities on Leafy and Fruiting Crops in Aquaponic Hoophouse Production

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**Abstract.** Aquaponics, the practice of growing fish and plants in the same system, has become increasingly popular as a solution to food scarcity, especially in urban areas. However, most current research is focused on fish rearing or growth of plants. Our research focused on planting density for two leafy plants, kale (*Brassica oleracea* L.) and cilantro (*Coriandrum sativum* L.), and two fruiting plants, banana peppers (*Capsicum annuum* L.) and pole beans (*Phaseolus vulgaris* L.), at high (30, 37, 12, and 14 plants/m<sup>2</sup>) and low (20, 26, 6, and 7 plants/m<sup>2</sup>) planting densities, respectively. Six aquaponic systems were set up in a hoophouse in Stillwater, OK, USA, and stocked with bluegill (*Lepomis macrochirus* L.) fingerlings. All crops were found to have a greater vegetative biomass at lower planting densities, as well as greater chlorophyll content in all plants except banana peppers. Number and weight of fruit did not change significantly between planting densities, but peppers were longer at greater densities.

Modern problems often require modern solutions, but lack of land, or degradation of previously arable land, has transcended time as civilization has expanded (Právělie et al. 2021). Approximately 40% of all arable land is experiencing some form of degradation, usually via soil erosion, with urban centers constantly expanding and reducing agricultural land (Právělie et al. 2021; Shi et al. 2016). Urban expansion into rural areas, also known as urban creep, has been increasing in recent years as urban populations rise and developing nations grow (Mohammadi et al. 2021; Pandey and Seto 2015; Parsipour et al. 2019). Urban creep affects most countries, progressing more rapidly in developing nations, and requires solutions to protect agricultural land and integrate agriculture into urban areas (Parsipour et al. 2019). As more than 309 million people globally are facing acute food scarcity, integrating agriculture into urban landscapes has become more important than ever (World Bank 2022; World Food Programme 2023). Aquaponics, the combination of aquaculture and hydroponics, may be part of the integration solution.

Use of aquaponic systems dates to 1000 A.D., when the Aztecs conquered their lack of land by covering rafts in soil, planting crops, and suspending the rafts in ponds and lakes (Jones 2002; Okomoda et al. 2023). Modern aquaponic systems can be used in many locations because of their low land requirements beyond electricity and limited

fresh water, especially in “unusable” potential agricultural landscapes, such as urban areas and poor agricultural land (El-Essawy et al. 2019). Although aquaponic systems have been used previously, primarily in greenhouse settings, use of these systems without a controlled environment is possible in many locations, such as outdoors in favorable climates, hoophouses, and rooftops (Love et al. 2015). In a study using a geographic information system-based multi-criteria decision analysis to determine ideal location for aquaponics utilization, Zaniboni et al. (2024) found that the most favorable locations for aquaponics were around principal cities, suggesting that aquaponics could be incorporated into urban creep.

In addition, unlike in traditional field agriculture and many hydroponic growing systems, aquaponic systems use a closed recirculating system, producing minimal amounts of wastewater and promoting long-term sustainability and limited runoff in urban areas. In fact, aquaponic systems may improve water use by 85% and produce 100% to 150% more plant matter compared with traditional field agriculture (AlShrouf 2017). Not only can aquaponic systems produce all-organic, nutritious leafy greens and fruit, but also they produce fish, a commonly consumed protein by most individuals and cultures, suiting the diversity found in urban centers (Clark and Tilman 2017). However, scant research has been done to improve grower recommendations for plants in aquaponics, leaving a dearth of knowledge for prospective business owners and growers.

One such realm is planting density for various types of aquaponic crops. Planting density, the number and spacing of plants in a specified area, is as important as fish stocking rate, as plants remove the nitrates from the cycling water, purifying the water for the aquatic species

used (Boxman et al. 2017). Most planting densities found in the literature are based on personal experience, traditional field agricultural standards, or fish stocking rate (Pickens et al. 2016; Somerville et al. 2014; Sumar et al. 2015). However, as a result of the increases in nutrients provided by fish waste fertilizer, the possibility of increasing planting density may provide a novel opportunity to produce more crops in less space.

Current recommendations are conflicting, as each experiment uses a different planting density. Sumar et al. (2015), citing experience, suggested that 12 plants/m<sup>2</sup> was the appropriate density for spinach (*Spinacia oleracea* L.) and tomato (*Solanum lycopersicum* L.) in a 6 m<sup>2</sup> grow bed, whereas Pickens et al. (2016) recommended densities of 24.2 plants/m<sup>2</sup> for leafy crops and 4 plants/m<sup>2</sup> for vining crops. Conversely, Afolabi (2020) stocked kale at density of 25 plants/m<sup>2</sup> in a comparison study of deep-water culture vs. integrated aquavegeticulture, and Ani et al. (2022) used 16 plants/m<sup>2</sup> for a fish density trial using lettuce (*Lactuca sativa* L.) as the representative plant. Baßmann et al. (2020) found that greater densities of basil influenced African catfish (*Clarias gariepinus* L.) welfare and behavior positively, whereas greater densities (144 plants) of basil (*Ocimum basilicum* L.) had 1.7% less leaf area than lower densities (n = 48) in 1.5 m<sup>2</sup> grow areas, but Boxman et al. (2017) found that planting density did not affect plant growth in a marine aquaponic system. In a technical article by Somerville et al. (2014), densities for 12 different plants are given; however, there is no reasoning given behind the recommendations. No commonly accepted standards for planting density in aquaponic grow beds exist, leaving those interested in aquaponic systems to guess at guidelines for planting. This study sought to examine two densities of several vegetable and herb species to provide recommendations of plant density in aquaponic systems.

## Materials and Methods

This experiment took place at the Botanic Garden at Oklahoma State University in Stillwater, OK, USA, in a hoophouse on the premises. Six identical aquaponic systems (20 EXT Aquaponic System; Symbiotic Aquaponic, McAlester, OK, USA) were purchased. Each system had a 492.1 L fish tank, 1.9 m<sup>2</sup> media bed grow space, and a 30 L·min<sup>-1</sup> flow rate. Tanks were stocked with 45 bluegill (*Lepomis macrochirus* L.) fingerlings (Moore’s Fish Farm, Inola, OK, USA) each, and fish were replaced or culled as needed to correct for death and illness. After 8 months of growth, bluegill fish were culled to half their population to prevent overcrowding. Bluegill fish were weighed at the beginning and end of each plant growth cycle, and fish reaching 0.45 kg were harvested at the end of each cycle. Fish were fed to satiation with 1.5 mm floating pellets with 40% crude protein (Optimal Fish Food, Omaha,

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NE, USA) two to four times a day (John and Mahalingam 2021) based on age.

Four species of plants were evaluated for crop density in these systems: kale (*Brassica oleracea* var. *acephala* L. 'Winterbor'), cilantro (*Coriandrum sativum* L. 'Cruiser'), banana pepper (*Capsicum annuum* L. var. *Goddess* F1), and pole beans (*Phaseolus vulgaris* L. 'Seychelles OG') (Johnny's Seeds, Winslow, ME, USA). Two densities of planting were chosen, with high densities being comprised of double the field planting recommendations with 54 kale plants, 68 cilantro plants, 14 pepper plants, and 22 pole bean plants (30, 37, 12, and 14 plants/m<sup>2</sup> respectively), whereas low densities were comprised of current field planting recommendations with 36 kale plants, 48 cilantro plants, 7 pepper plants, and 11 pole bean plants (20, 26, 6, and 7 plants/m<sup>2</sup> respectively). Kale and cilantro cultivars were planted in 2.5 cm rockwool cubes, whereas peppers and beans were planted in 3.8 cm rockwool cubes (Grodan, Roermond, Netherlands).

Each density of plants was repeated in two beds per replication for a total of two repetitions for each plant species per replication. Each plant species was planted sequentially for a total of two replications over time per plant species. Kale and cilantro were grown over winter in the first year, and pepper and beans were grown over the summers of the first and second years of growing.

The average temperature during the course of the experiment was  $10.4 \pm 8.2$  and  $20.5 \pm 12.9^\circ\text{C}$  for kale,  $23.6 \pm 14.0$  and  $25.0 \pm 11.7^\circ\text{C}$  for cilantro,  $31.8 \pm 9.5$  and  $25.2 \pm 10.6^\circ\text{C}$  for peppers, and  $17.9 \pm 9.2$  and  $21.6 \pm 11.7^\circ\text{C}$  for beans. Relative humidity averaged  $34.9 \pm 15.5$  and  $49.9 \pm 15.6\%$  for kale,  $35.5 \pm 20.3$  and  $43.6 \pm 23.1\%$  for cilantro,  $40.9 \pm 21.5$  and  $37.3 \pm 20.7\%$  for peppers, and  $46.0 \pm 23.5$  and  $26.7 \pm 24.4\%$  for beans. Daily light integrals averaged  $5.7 \pm 7.6$  and  $11.2 \pm 19.7 \text{ mol}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$  for kale,  $24.0 \pm 25.3$  and  $18.6 \pm 21.6 \text{ mol}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$  for cilantro,  $20.6 \pm 14.6$  and  $21.3 \pm 18.3 \text{ mol}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$  for peppers, and  $17.3 \pm 27.0$  and  $18.6 \pm 23.1 \text{ mol}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$  for beans.

**Data collection.** Water pH, electrical conductivity (EC), and temperature were measured daily using a portable pH/EC/temperature meter (Hanna Instruments, Smithfield, RI, USA). Ammonia, nitrite, and nitrate levels were measured weekly (API Test Kit; Aquarium Pharmaceuticals Inc. McLean, VA, USA). All water quality parameters were kept within recommended ranges using water changes, pH up powder (Symbiotic Aquaponic), and General Hydroponics pH Down Liquid (Hawthorne Hydroponics, Tyler, TX, USA) (Tables 1 and 2, Figs. 1–3). Supplemental chelated iron (Symbiotic Aquaponic) was added as needed.

Height, dry weight, and total dry root biomass were taken for each plant after 7 weeks of growth for kale and 6 weeks of growth for cilantro. Height was taken on four representative plants each week during the course of the growing cycle. In addition, chlorophyll was measured using a soil plant analysis development (SPAD) meter (SPAD-502; Konica

Table 1. Nutrients found in water samples taken at the end of each growing cycle for two densities grown in aquaponic systems in a hoop-house in Stillwater, OK, USA.

Nutrient (ppm)	Plant density		<i>P</i> value <sup>i</sup>
	High	Low	
Sodium	170.3 a <sup>ii</sup>	194.3 a	0.5737
Potassium	25.6 a	21.9 a	0.5238
Calcium	50.6 a	51.2 a	0.8615
Magnesium	48.3 a	49.9 a	0.7509
Chloride	161.8 a	186.1 a	0.5141
Boron	0.36 a	0.39 a	0.5690
Nitrate-nitrogen	17.2 a	15.8 a	0.7901
Sulfate	230.8 a	255.5 a	0.6344
Bicarbonate	143.5 a	139.6 a	0.8776
Zinc	0.8 a	0.8 a	0.9178
Iron	1.8 a	2.1 a	0.6026

<sup>i</sup>All *P* values are nonsignificant.

<sup>ii</sup>Means within a row followed by same lowercase letter are not significantly different by Tukey's comparison in linear mixed models ( $P \leq 0.05$ ).

Minolta, Tokyo, Japan) at the end of the growing period. Two SPAD readings were taken from each kale and cilantro plant, one from an inner leaf and one from an outer leaf. Readings were averaged to determine the chlorophyll concentration. Fresh foliar rate and Brix readings, using a handheld refractometer (Sungrow, Carrollton, TX, USA), were taken at the end of the growth period for kale and cilantro. Shoot and root samples were dried at  $59^\circ\text{C}$  for 1 week.

Height, fresh weight, dry weight, and total dry root weight were taken for each plant after 13 weeks of growth for peppers and 10 weeks of growth for beans. Fruit were harvested, weighed, and measured each week for 5 weeks for peppers and beans, and Brix measurements were taken for each fruit harvested using the refractometer. In addition, chlorophyll was measured using the SPAD meter at the end of the growing period. Three SPAD readings were taken from each pepper and bean plant on a top, middle, and bottom leaf. Readings were averaged to determine the chlorophyll concentration. Shoot and root samples were dried at  $59^\circ\text{C}$  for 1 week.

Three dried samples comprised of four plants each were chosen randomly to be analyzed for nutrients. Water samples were collected from the fish tank, sump tank, and grow bed after each replication. Both the dried samples and water samples were then

sent to the Soil, Water and Forage Analytical Laboratory at Oklahoma State University (Stillwater, OK, USA) for analysis of leaf mineral element concentrations and water nutrients using a nutrient analyzer (TruSpec Carbon and Nitrogen Analyzer; LECO Corp., St. Joseph, MI, USA).

**Statistical analysis.** The study was a randomized complete block design with two replications per plant species, blocking for repetition and planting date. Water quality data were treated as a randomized complete block design with two replications per plant species and were blocked for repetition to compare water quality at differing densities. Statistical analysis was performed using SAS/STAT software (version 9.4; SAS Institute, Cary, NC, USA). Tests of significance were reported at the 0.05 level. Data were analyzed using generalized linear mixed-models methods, with Tukey's multiple comparison methods used to separate the means.

## Results and Discussion

**Water quality parameters.** No significant differences in water quality parameters were observed between the two planting densities for any crop (Tables 1–3, Figs. 1 and 2).

**Planting density effects on plant growth.** All crops showed an increase in individual plant biomass at low densities compared with high densities (Table 4). Bennett et al. (2013) found that lower planting densities (6.7 plants/m<sup>2</sup> compared with 10 and 25 plants/m<sup>2</sup>) impacted overall vegetative growth positively as a result of a reduction in resource competition. Naiki and Gupta (2010) found that lower planting density and more space between plants increased leaf weight and overall plant, but not yield per plot, and Singh et al. (2003) found similar results in kohlrabi (*Brassica oleracea* var. *gongylodes* L.). Although the stem diameter and stem-to-leaf ratio were greater as space between plants increased, overall yield tended to decrease as spacing increased (Patil et al. 2003; Singh et al. 2003). Nasto et al. (2009) found that pepper (*C. annuum* L. var. *Gogozhare*) plants had greater fresh weight at 11.1 plants/m<sup>2</sup> compared with 2.6 plants/m<sup>2</sup>. Likewise, Ahmad et al. (2021) found that denser planting (60 × 30 cm) of sweet banana pepper (*C. annuum* L.) increased fresh weight, and therefore dry weight, of pepper plants.

Table 2. Water quality properties found in samples taken at the end of each growing cycle for two densities grown in aquaponic systems in a hoop-house in Stillwater, OK, USA.

Water quality property	Plant density		<i>P</i> value <sup>i</sup>
	High	Low	
Total dissolved solids (ppm)	935.2 a <sup>ii</sup>	992.4 a	0.6653
Potassium absorption ratio	0.36 a	0.31 a	0.5167
Sodium absorption ratio	4.0 a	4.5 a	0.5767
Exchangeable potassium (%)	6.9 a	6.4 a	0.5029
Exchangeable sodium (%)	4.4 a	4.9 a	0.5941
Hardness (ppm)	324.1 a	332.8 a	0.7573
Alkalinity (ppm)	119.2 a	115.4 a	0.8629

<sup>i</sup>All *P* values are nonsignificant.

<sup>ii</sup>Means within a row followed by the same lowercase letter are not significantly different by Tukey's comparison in linear mixed models ( $P \leq 0.05$ ).

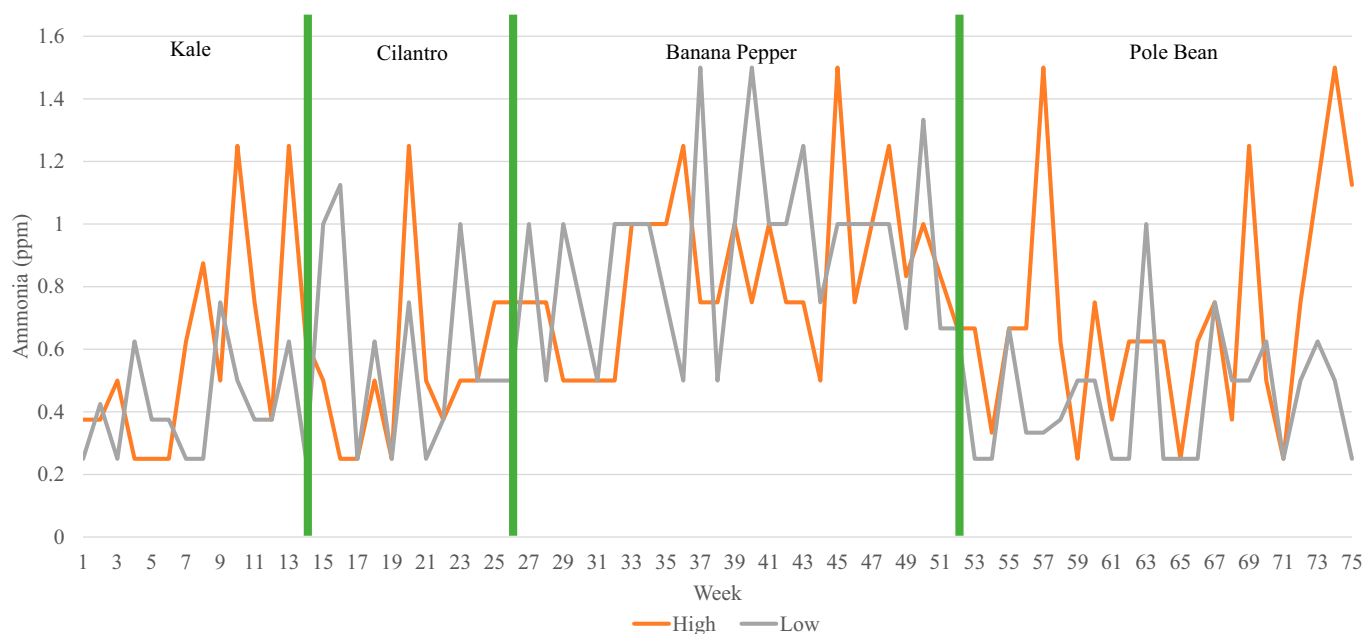


Fig. 1. Weekly average ammonia levels in aquaponic systems planted with two different densities of four crops (kale, cilantro, banana peppers, and pole beans) over time in a hoophouse in Stillwater, OK, USA.

Both cilantro and pepper plants were taller at greater planting densities than at lower planting densities (Table 4). Ahmad et al. (2021) found that denser planting ( $60 \times 30$  cm) of sweet banana pepper produced taller plants than the  $60 \times 60$  cm spacing, as did Nasto et al. (2009). Conversely, Aminifard et al. (2012) observed that sweet pepper height decreased with increased planting density. Paprika peppers also decreased in height as plant spacing decreased (Aminifard et al. 2010). Conversely, many previous studies on planting density in cilantro showed no significant effect or a decrease in height at greater planting densities (Kahn and Maness 2010; Li et al. 2024; Painkara et al. 2024). However,

Mangan et al. (2001) found that cilantro planted at greater densities increased in height, similar to our study. Although competition for nutrients can decrease growth, competition for light can cause plant plasticity, leading to more allocations for plant growth to out-shade nearby competition (Xiao et al. 2006).

Kale, cilantro, and pole beans had greater SPAD values, leading to a greater chlorophyll content at the lower planting density (Table 4). Although in some plant species, such as certain varieties of rice (*Oryza sativa* L.), plant density does not affect chlorophyll content, both kale and cilantro have been shown previously to experience an increase in chlorophyll content as plant spacing increases (Arjona 1980; Mondal

et al. 2013; Naiki and Gupta 2010; Sharma et al. 2016). Arjona (1980) found that, as kale was spaced from 22 to 30 cm, chlorophyll content increased significantly, whereas additional spacing did not increase chlorophyll content. Noboa et al. (2022) measured comparable SPAD values, as increasing hydroponic row spacing allowed for an increase in chlorophyll content. Similarly, Naiki and Gupta (2010) found that increasing spacing from  $30 \times 20$  cm to  $30 \times 40$  cm produced kale with a greater chlorophyll content. Sharma et al. (2016) found that increasing spacing, thus decreasing planting density, improved yield and other quality parameters, such as chlorophyll content, in cilantro, likely as a result of increased space

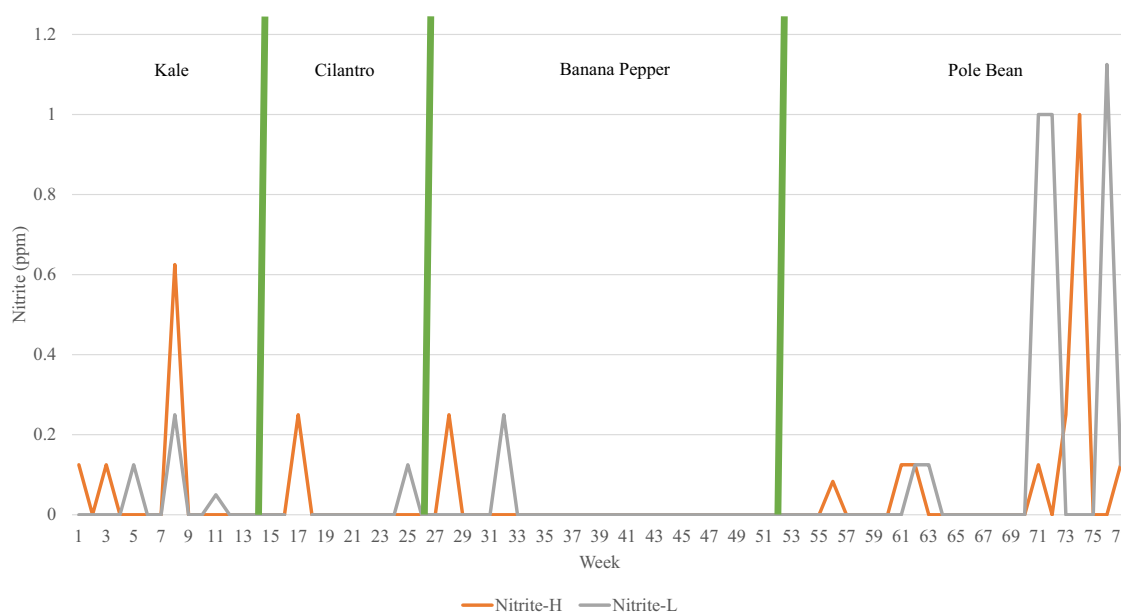


Fig. 2. Weekly average nitrite levels in aquaponic systems planted with two different densities of four crops (kale, cilantro, banana peppers, and pole beans) over time in a hoophouse in Stillwater, OK, USA.

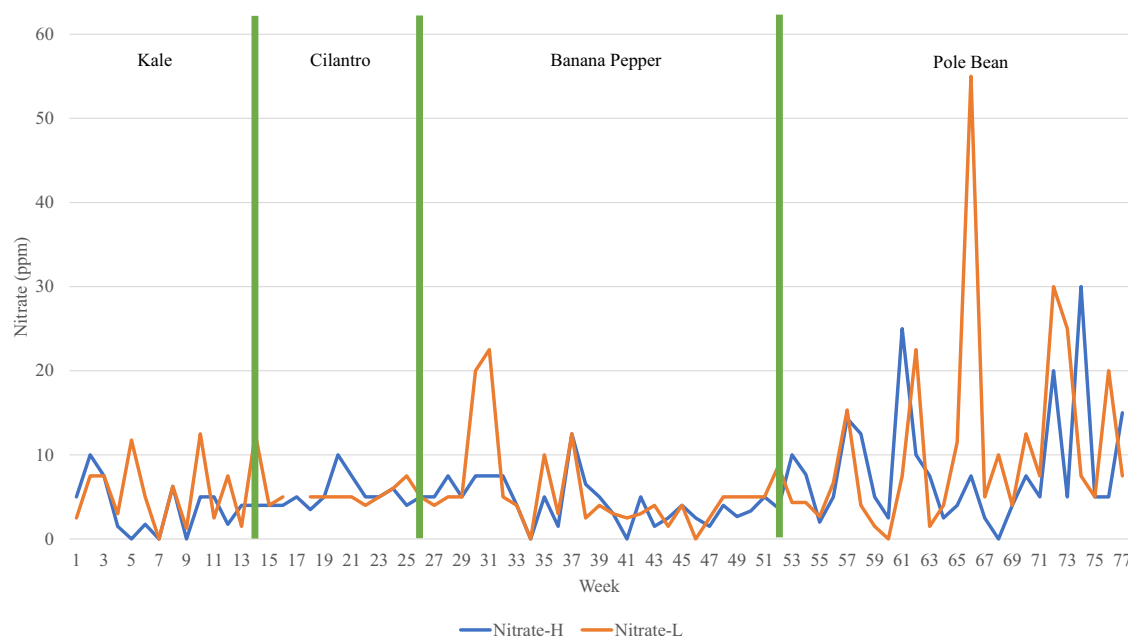


Fig. 3. Weekly average nitrate levels in aquaponic systems planted with two different densities of four crops (kale, cilantro, banana peppers, and pole beans) over time in a hoophouse in Stillwater, OK, USA.

Table 3. Weekly water quality parameters taken on aquaponic systems planted with different densities over the course of eight growing cycles in Stillwater, OK, USA.

Water quality parameter	Plant density		Recommended range	P value <sup>i</sup>
	High	Low		
Daily temperature (°C)	22.2 a <sup>ii</sup>	21.7 a	26–32 <sup>iii</sup>	0.0727
Daily pH	7.9 a	7.9 a	6.5–8.5	0.4566
Daily electrical conductivity (ms·cm <sup>-1</sup> )	0.9 a	1.0 a	0.205 <sup>iv</sup>	0.3125
Weekly ammonia (ppm)	0.7 a	0.6 a	<8.0	0.1443
Weekly nitrite (ppm)	0.0 a	0.1 a	<1.0 <sup>v</sup>	0.6044
Weekly nitrate (ppm)	5.8 a	7.4 a	<250.0	0.1626

<sup>i</sup> All P values are nonsignificant.

<sup>ii</sup> Means within a row followed by the same lowercase letter are not significantly different by Tukey's comparison in linear mixed models ( $P \leq 0.05$ ).

<sup>iii</sup> Recommended ranges for temperature, pH, ammonia, nitrite, and nitrate derived from Akinwale and Adeola (2012).

<sup>iv</sup> Recommended range for electrical conductivity by Stone et al. (2013).

<sup>v</sup> Recommended range for nitrite by Bhatnagar and Pooja (2013).

allowing for abundant vegetative control. Increases in chlorophyll content with lower planting density are likely a result of shading competition in tightly spaced plants (Arjona 1980). In addition, increases in chlorophyll content may also be the result of a greater availability of water, nutrients, and sunlight, and an overall reduction in competition (Naiki and Gupta 2010).

*Planting density effects on nutrient content of kale, pepper, and bean plants.* Although most secondary and micronutrients were not found to be significantly different in any plant species at either planting density, kale showed a significant difference in sulfur content between the high and low planting densities (Table 5). *Brassica* species are known for high levels of secondary sulfur compounds,

Table 4. Main effects of planting density on the physical characteristics of four crops in aquaponic systems stocked with bluegill in Stillwater, OK, USA.

Plant species and density	Fresh shoot wt (g)	Dry shoot wt (g)	SPAD	Height (cm)	Total root biomass (g)	°Brix
<b>Kale</b>						
30 plants/m <sup>2</sup>	71.8 b <sup>i</sup>	6.8 b	42.7 b	25.8 a	193.0 a	7.1 a
20 plants/m <sup>2</sup>	90.5 a	8.5 a	44.3 a	26.0 a	166.2 a	6.7 a
P value	0.0031**	0.0069**	0.0044**	0.7211 <sup>NS</sup>	0.6138 <sup>NS</sup>	0.0620 <sup>NS</sup>
<b>Cilantro</b>						
37 plants/m <sup>2</sup>	59.3 b	4.3 a	37.1 b	31.1 a	663.3 a	4.2 a
26 plants/m <sup>2</sup>	69.8 a	5.6 b	38.7 a	29.6 b	685.8 a	4.5 a
P value	0.0173*	0.0004***	<0.0001***	0.0330*	0.9371 <sup>NS</sup>	0.0848 <sup>NS</sup>
<b>Pepper</b>						
8 plants/m <sup>2</sup>	— <sup>ii</sup>	100.2 b	50.8 a	101.2 a	912.6 a	—
4 plants/m <sup>2</sup>	—	127.4 a	52.7 a	91.3 b	353.2 a	—
P value	—	0.0139**	0.1023 <sup>NS</sup>	0.0477*	0.3136 <sup>NS</sup>	—
<b>Bean</b>						
12 plants/m <sup>2</sup>	—	71.3 b	38.8 b	179.8 a	147.3 a	—
6 plants/m <sup>2</sup>	—	110.4 a	41.7 a	201.9 a	547.2 a	—
P value	—	0.0309*	0.0068**	0.1137 <sup>NS</sup>	0.1944 <sup>NS</sup>	—

<sup>i</sup> Means within a column followed by the same lowercase letter are not significantly different by Tukey's comparison in linear mixed models ( $P \leq 0.05$ ).

<sup>ii</sup> Data not collected on nonedible plant parts.

NS, \*, \*\*, \*\*\* Nonsignificant or significant at  $P \leq 0.05$ ,  $P \leq 0.01$ , or  $P \leq 0.001$ , respectively.

SPAD = soil plant analysis development.

Table 5. Foliar nutrient contents of four crops grown in aquaponic systems in Stillwater, OK, USA at two different planting densities.

Plant species and density	Total nitrogen (%)	Phosphorus (%)	Potassium (%)	Magnesium (%)	Calcium (%)	Sulfur (%)
Kale						
30 plants/m <sup>2</sup>	5.4 a <sup>i</sup>	0.7 a	4.6 a	0.6 a	3.0 a	1.3 a
20 plants/m <sup>2</sup>	5.6 a	0.7 a	4.8 a	0.5 a	3.0 a	1.2 b
P value	0.2308 <sup>NS</sup>	0.2469 <sup>NS</sup>	0.1869 <sup>NS</sup>	0.1065 <sup>NS</sup>	0.7322 <sup>NS</sup>	0.0025***
Cilantro						
37 plants/m <sup>2</sup>	5.3 a	1.1 a	8.9 a	0.6 a	1.4 a	0.3 a
26 plants/m <sup>2</sup>	5.3 a	1.1 a	8.8 a	0.6 a	1.4 a	0.3 a
P value	0.7369 <sup>NS</sup>	0.3700 <sup>NS</sup>	0.8902 <sup>NS</sup>	0.2620 <sup>NS</sup>	0.6398 <sup>NS</sup>	0.8677 <sup>NS</sup>
Pepper						
8 plants/m <sup>2</sup>	3.9 a	0.4 a	6.5 a	1.0 a	1.3 a	0.5 a
4 plants/m <sup>2</sup>	4.0 a	0.4 a	5.5 b	1.0 a	1.4 a	0.5 a
P value	0.5392 <sup>NS</sup>	0.4003 <sup>NS</sup>	0.0341*	0.8744 <sup>NS</sup>	0.2799 <sup>NS</sup>	0.5716 <sup>NS</sup>
Bean						
12 plants/m <sup>2</sup>	2.4 b	0.5 a	2.9 b	0.9 a	2.0 a	0.2 b
6 plants/m <sup>2</sup>	2.9 a	0.5 a	3.3 a	1.0 a	2.0 a	0.2 a
P value	0.0163**	0.5612 <sup>NS</sup>	0.0454*	0.5326 <sup>NS</sup>	0.9033 <sup>NS</sup>	0.0192**

<sup>i</sup> Means within a column followed by the same lowercase letter are not significantly different by Tukey's comparison in linear mixed models ( $P \leq 0.05$ ).

NS, \*, \*\*, \*\*\* Nonsignificant or significant at  $P \leq 0.05$ ,  $P \leq 0.01$ , or  $P \leq 0.001$ , respectively.

such as glucosinolates (Aghajanzadehdivaei 2015). Nasiri et al. (2017) found that canola (*Brassica napus* L.) cultivars increased in glucosinolates as planting density increased from 40 to 80 plants/m<sup>2</sup>. Similarly, Phong et al. (2019) found that glucosinolates were greatest in watercress (*Nasturtium officinale* L.; Brassicaceae) on day 35 of growth when plants were spaced at 14 cm compared with 20 and 30 cm planting densities. Chishaki and Horiguchi (1997) found that in many plants, such as rice and barley, nutrient deficiencies caused an increase in secondary metabolites. As plant density increases, competition for resources, such as light and nutrients, increases. Competition among plants increases stress, which can increase secondary metabolites such as glucosinolates (Phong et al. 2019). In addition, Koralewska et al. (2007) found that sulfur deprivation increased sulfur uptake 4-fold compared with sulfate-rich environments, suggesting that a lack of sulfur and sulfates resulting from competition could increase sulfur uptake and storage.

Pepper plants increased in potassium at a high density, whereas beans had increased potassium at a low density (Table 5). Many studies do not report on the leaf mineral composition of fruiting crops under various planting densities, thus leaving a dearth of information. Rajaraman and Pugalandhi (2013) reported that okra (*Abelmoschus esculentus* L. var. Moench) leaves had a greater potassium content at

lower planting densities. Conversely, Matsoukis et al. (2015) found that shading increased potassium content in West Indian lantana (*Lantana camara* L.). Menzel and Simpson (1988) found that shading did not affect the foliar potassium content in passion-fruit (*Passiflora edulis* f. *edulis* × *P. edulis* f. *flavicarpa* hybrid, E-23), whereas iron, calcium, and magnesium increased, and phosphorus declined.

*Planting density effect on pepper fruit length.* At the greater planting density (8 plants/m<sup>2</sup>), pepper plants produced longer fruit compared with the low planting density (4 plants/m<sup>2</sup>) (Table 6). These findings are unusual compared with past research. Kim et al. (1999) and Daşgan and Abak (2003) found no significant difference in pepper size at increased planting densities, whereas Islam et al. (2018) found that pepper fruit length decreased as plants were spaced more closely. Nasto et al. (2009) found that peppers planted at densities of 4.8 plants/m<sup>2</sup> and less had a greater fresh weight and diameter compared with higher densities. Aminifard et al. (2012) concurred, finding that closer spacing produced less and smaller fruit than wider spacing. However, plots spaced at 20 × 50 cm and supplemented with nitrogen fertilizer at 100 kg/ha<sup>-1</sup> did not produce fruit significantly different from plots spaced at 20 × 100 and 30 × 100 cm supplemented with less nitrogen, suggesting that addition of nitrogen has the potential to

override the reducing effects of crowding (Aminifard et al. 2012). In addition, the heat of the hoophouse in our experiment may have affected fruit growth and development, as high temperatures can cause flower drop and reduced growth (Pagamas and Nawata 2008; Thuy and Kenji 2015).

## Conclusion

Plants with larger leaves, such as kale, require increased growing space, whereas herbs, such as cilantro, do not. Fruit plants, such as banana peppers and pole beans, may thrive under crowded conditions, especially in aquaponics, as nitrogen is usually in excess. Research on planting density of several different categories of plants in aquaponics has not been done previously with both leafy greens and fruit.

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Table 6. Physical characteristics and approximate sugar content of banana pepper and pole bean fruit at two planting densities in aquaponic systems set up in a hoophouse in Stillwater, OK, USA.

Plant species and density	Total no. of fruit	Total fresh wt (g)	Avg individual fresh wt (g)	Length (cm)	°Brix
Pepper					
8 plants/m <sup>2</sup>	165.0 a <sup>i</sup>	2464.1 a	23.4 a	12.4 a	5.6 a
4 plants/m <sup>2</sup>	115.5 a	3872.2 a	21.3 a	11.1 b	5.8 a
P value	0.1898 <sup>NS</sup>	0.1208 <sup>NS</sup>	0.0657 <sup>NS</sup>	<0.0001***	0.5150 <sup>NS</sup>
Bean					
12 plants/m <sup>2</sup>	165.0 a	5218.3 a	6.2 a	12.1 a	6.4 a
6 plants/m <sup>2</sup>	115.5 a	3894.1 a	5.4 a	12.2 a	6.3 a
P value	0.1898 <sup>NS</sup>	0.1208 <sup>NS</sup>	0.2129 <sup>NS</sup>	0.4443 <sup>NS</sup>	0.5162 <sup>NS</sup>

<sup>i</sup> Means within a column followed by the same lowercase letter are not significantly different by Tukey's comparison in linear mixed models ( $P \leq 0.05$ ).

NS, \*\*\* Nonsignificant or significant at  $P \leq 0.001$ , respectively.

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