HortScience 60(3):419–425. 2025. https://doi.org/10.21273/HORTSCI18393-24

Impact of Seasonal Temperature Changes on Sesquiterpene Lactone and Sugar Concentrations in Hydroponically Grown Lettuce

Mason McLemore, Shivani Kathi, John Unterschuetz, Tyler Mason, Niels Maness, Donna Chrz, Bruce Dunn, Charles Fontanier, and Bizhen Hu

Department of Horticulture and Landscape Architecture, 320G Agriculture Hall, Oklahoma State University, Stillwater, OK 74078, USA

Keywords. bitterness, fructose, glucose, *Lactuca sativa*, lactucopicrin, quality, yield

Abstract. **Lettuce is one of the most important leafy vegetables in the United States, and it is subjected to a decrease in edible quality when cultivated in environments with hot temperatures and increased daylength. Specifically, bitter-tasting compounds called sesquiterpene lactones (SLs) accumulate in certain cultivars and lettuce market types, especially during the bolting stage. However, increased sugar accumulation may offset bitterness, with high sugar:SL ratios reducing the bitter flavor perception. Hence, it is important to determine the effect of seasonal temperatures on sugar, SL, and sugar:SL accumulation in lettuce cultivars to predict the likelihood of perceived bitterness. Twelve cultivars spread across four different commonly grown market types (i.e., Romaine, Butterhead, Batavian, and Salanova®) were grown for four harvest seasons (Spring 2020, Fall 2020, early Summer 2021, and Winter 2021) in nutrient flow technique hydroponic culture. Lettuces exhibited significant differences in harvest seasons, cultivar, and their interaction for free SLs and total SLs, sucrose, and sugar:SL ratio. Specifically, plant fresh weight and total SLs were greatest in Spring 2020 and Fall 2020, respectively, and lowest in Winter 2021. Total sugars were the same between harvests, and Winter 2021 had a significantly lower sugar:SL ratio than that of the other three harvests. Cultivars included in this study within romaine (Parris Island, Jericho, and Coastal Star) and Batavian market types (Nevada and Sierra, but excluding Cherokee) emerged as top candidates to grow during the summer heat because of the higher plant weights, sugar concentrations, and lower sugar:SL ratio, as well as decreased SL concentrations. Overall, SL and sugar concentrations were notably low in the Winter 2021 harvest season for all cultivars and market types. Growers can optimize their production and ensure better plant yield and quality by strategically choosing lettuce market types and cultivars that have better plant growth and predicted flavor for different seasons in a greenhouse.**

Lettuce (*Lactuca sativa* L.) is one of the most significant leafy green vegetable crops cultivated in the United States, accounts for approximately $4.4 billion received in cash receipts from its sales, and ranks as the most widely consumed leafy green in the United States (US Department of Agriculture 2023). Adding to its popularity, lettuce encompasses a diverse array of market types such as looseleaf, romaine, butterhead, Batavian, and proprietary types like Salanova®. Each of these types exhibits unique morphological and genetic traits, such as variations in structure, midrib formation, and levels of heat tolerance. Adaptability to heat is an increasingly crucial characteristic in regions with hot climates, such as Oklahoma (Thakulla et al. 2021). Research has shown that heat tolerance varies significantly among cultivars. In addition, certain market types yield greater fresh weights under elevated temperatures. For example, cultivars like Nevada and Parris Island have demonstrated higher yield in high-temperature greenhouse environments compared with that of others such as Buttercrunch and Coastal Star (Holmes et al. 2019).

Lettuce is a cool season crop that prematurely bolts under heat stress, especially under long light durations. Bolting is characterized by the transition from vegetative to reproductive growth (Hao et al. 2018), which leads to the redistribution of sugars from leaves to reproductive tissues (Khan 2018; Lee and Sugiyama 2006). Further, this transition triggers the accumulation of bitter-tasting compounds known as sesquiterpene lactones (SLs), thereby negatively affecting edible quality. Sugar and SL concentrations interact with each other, and higher concentrations of sugar and lower concentrations of SLs are associated with an appealing and nonbitter flavor perception. This highlights the importance of the sugar:SL ratio in determining overall lettuce quality (Chadwick et al. 2016).

Lettuce quality is influenced by seasonal production factors, including maximum daily temperature, light intensity and especially light duration, and humidity, which fluctuate throughout the year (Sublett et al. 2018). Greenhouse cultivation offers a potential solution to mitigate these seasonal effects by providing a controlled environment that moderates temperature variations. Lettuce production in greenhouses is gaining popularity worldwide because of its higher than average field produced yields, year-round production availability, and improved crop management practices (Gargaro et al. 2023). Recent studies have demonstrated that controlled environment agriculture can significantly enhance crop quality and yield. Specifically, lettuce grown in a climate-controlled setting exhibited reduced bolting rates and improved taste profiles compared with those of lettuce grown outdoors (Hernandez et al. 2020; Wang et al. 2023). Greenhouse mechanical systems help regulate high summer temperatures and low winter temperatures, facilitating year-round production (Lei and Engeseth 2021). However, seasonal microclimate differences in terms of temperatures and humidity still exist within greenhouses which could have a significant influence on plant performance, yield, and edible quality; however, their severity, depends on the greenhouse design, outside climate, and technology available to control the environmental conditions within the greenhouse, such as heating, cooling, and ventilation systems (Šalagovič et al. 2024). Furthermore, a study by Li et al. (2023) highlighted the genetic variability among lettuce cultivars in response to environmental stresses, thus emphasizing the need for cultivar selections that consider both seasonal variation as well as macroclimate or microclimate changes. Hence, it is important to select the right cultivars suitable for different seasonal variations within a greenhouse. However, there is little literature regarding the effect on seasonal variations in greenhouses on lettuce quality in terms of sugars, SL, and sugar: SL ratio.

This study aimed to investigate the effects of different seasons on specific lettuce cultivars grown in a nutrient flow technique (NFT) greenhouse production environment. Understanding the responses of cultivars to changing greenhouse environmental conditions throughout the year can enhance production strategies and improve the quality of lettuce. Hence, the current study investigated the extent to which plant weight, SL, sugar, and sugar:SL ratio in hydroponically grown lettuce were influenced by seasonal variation and cultivars within various market types. The objective was to evaluate yield, expressed as fresh plant weight, and quality indicators (SL, sugar, and sugar:SL ratio) of 12 different lettuce cultivars grown hydroponically across four different seasons (i.e., spring, fall, summer,

Received for publication 12 Dec 2024. Accepted for publication 20 Jan 2025.
Published online 26 Feb 2025.
S.K. is the corresponding author. E-mail: shivani.kathi@okstate.edu.
This is an open access article distributed under the CC BY-NC license (https://creativecommons.org/licenses/by-nc/4.0/).

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-16 via Open Access. This is an open access article distributed under the CC BY-NC license (https://creativecommons.org/licenses/by-nc/4.0/). https://creativecommons.org/licenses/by-nc/4.0/

and winter) of production. The overall goal of the study was to identify lettuce market types and cultivars that perform well in terms of yield and quality across seasons within a greenhouse environment.

Materials and Methods

Plant material. Twelve lettuce cultivars, including romaine, butterhead, Batavian, and Salanova® market types, were grown in this study. Seeds were purchased from Johnny's Selected Seed Company (Winslow, ME, USA) as pelleted or nonpelleted dependent on commercial availability of the pelleted form. Lettuce market type, cultivar, and seed form (pelleted or nonpelleted) information are included in Table 1. Seedlings were germinated in 1.5-cm^3 Oasis cubes (Oasis Grower Solutions, Kent, OH, USA) at a density of one plant per cube on a mist bench at the Greenhouse Learning Center, Oklahoma State University Campus (Stillwater, OK, USA; lat. 36.125769861756694°N, long. 97.07393133189352°W). Mist bench emitters were turned on in 10-min intervals for a duration of 5 s. Miracle Gro (200 ppm solution of 24N–4.8P–9.6K; Scotts Miracle-Gro Company, Marysville, OH, USA) was applied in a single application 2 weeks after placement on the mist bench. Seedlings were held on the mist bench for 4 weeks before transferring into hydroponic culture.

Hydroponic culture. Seedlings were transplanted into randomly assigned slots on Hydrocycle Pro NFT tables (Growers Supply, Dyersville, IA, USA) in a randomized complete block design with five plants per cultivar per replication and four replications, with a total of 20 plants per cultivar grown across different seasons (Spring 2020, Fall 2020, early Summer 2021, and Winter 2021) (Table 2).

Two NFT tables were used, and each table included 10 troughs with 18 planting holes per trough (180 planting holes per table). Each trough measured 10 cm wide × 5 cm deep × 900 cm long, with 20-cm spacing between planting holes. The tables had a decline in slope of approximately 3% between the inlet and drainage ends. Hydroponic solutions within each 150-L table reservoir were initially started at 1.0 mS electrical conductivity (EC) using fertilizer (Jack's Hydroponic Special 5N–12P–26K; JR Peters Inc., Allentown, PA, USA) and at pH 6.0 using pH down (General Hydroponics, Santa Rosa, CA, USA). The EC was gradually increased to 2.0 mS over the course of 2 weeks (0.5 mS EC/week). Solutions were then maintained at 2.0 mS EC by monitoring and adjusting daily. The EC and pH were measured using a dual EC/pH meter (HI 9831-6; Hanna Instruments, Woonsocket, RI, USA). The flowrate of nutrient solution was 1500 $L \cdot h^{-1}$ for each table. Dissolved oxygen was maintained between 8 and 14 ppm using an aquarium air pump (Hydrofarm; Active Aqua AAPA15L, Petaluma, CA, USA). The daily light integral (DLI), temperature, and humidity readings were recorded using a TR-7Ui multidata logger (T&D, Matsumoto City, Japan) (Fig. 1 and Table 2).

Table 1. Lettuce market type, cultivar[i], and seed form (pelleted or nonpelleted).

Type	Cultivar	Seed form[ii]
Romaine	Parris Island	NP
	Jericho	NP
	Coastal Star	P
Butterhead	Buttercrunch	NP
	Nancy	P
Batavian	Nevada	P
	Cherokee	P
	Sierra	NP
Salanova®	Butter Red	P
	Butter Green	P
	Sweet Crisp Red	P
	Sweet Crisp Green	P

[i] Seeds were purchased from Johnny's Selected Seed Company (Winslow, ME, USA).

[ii] Seed form: NP = nonpelleted; P = pelleted.

Table 2. Seeding, transplanting, harvest date, total days elapsedi, and average daily light interval for lettuce growth during each season at the Greenhouse Learning Center in Stillwater, OK, USA.

Season	Seeding date	Transplanting date	Harvest date	Total days elapsed[i]	Average DLI (mol)
Spring 2020	4 Mar 2020	30 Mar 2020 (26)[ii]	4 May 2020 (35)	61	15.4 ± 4.1
Fall 2020	29 Jul 2020	27 Aug 2020 (29)	28 Sep 2020 (32)	61	23.7 ± 3.2
Early Summer 2021	2 Jun 2021	7 Jul 2021 (35)	2 Aug 2021 (26)	61	24.1 ± 3.0
Winter 2021	14 Oct 2021	12 Nov 2021 (29)	20 Dec 2021 (38)	67	12.8 ± 2.6

[i] Total days elapsed since the seeding date.

[ii] Numbers in parentheses denote elapsed time since the previous stage.

Lettuce harvesting and processing. At harvest, the three (of five) representative lettuce plants based on overall appearance in each replication with adhering roots attached to the Oasis cube were lifted out of the hydroponic trough, placed in a labeled bag, transferred to laboratory facilities at the Noble Research Center on the Oklahoma State University campus, and held in a cold room at 2 °C before further processing on the same day. Lettuce plants were first cut at the Oasis cube, and the cube and roots were discarded. Damaged leaves were removed, and shoot fresh weights for each lettuce head were recorded as yield. Samples were washed, head cores were removed, and a final leaf weight was determined. Samples were labeled, secured in cheesecloth, placed in a freezer bag, and held in a walk-in freezer at −20 °C before freeze-drying. Samples were freeze-dried because it the most recommended method for quality retention (Calín-Sánchez et al. 2020) using a Harvest Right freeze-dryer (HRFD-PLrg-SS; Harvest Right, North Salt Lake, UT, USA) with a final shelf temperature of 21.1 °C, pressure at 23 kPa, and cold trap temperature of −40 °C for approximately 100 h. After completion of drying, lyophilized samples were weighed and ground into 120-mL brown bottles through a 1-mm screen using a UDY Cyclone Mill (UDY Corporation, Fort Collins, CO, USA). Immediately after grinding, duplicate samples of approximately 150 mg were weighed for each lettuce sample to undergo a moisture content analysis. Samples were placed in an oven at 80 °C for 48 h. Moisture content of freeze-dried samples was calculated as a percentage. The remainder of the sample was used for sugar and SL extraction and analyzed.

Sugar extraction and analysis. Sugar extraction and preparation for analysis were conducted according to Maness (2010) and Davies (1988), with some modifications. Approximately 200 mg of each freeze-dried sample was accurately weighed in duplicate 2-dram vials. Samples were extracted with 2 mL of 95% ethanol by boiling under reflux at 85 °C using a digital dry block heater (Isotemp; Fisher Scientific, Waltham, MA, USA) for 20 min, with mixing every 5 min. After extraction, samples were centrifuged for 15 min at 3000 g_n using a SpeedVac® centrifuge (SPD-121P; Thermo-Savant, Waltham, MA, USA) and filtered using Whatman 41 filter paper (Cole-Parmer, Vernon Hills, IL, USA) into 10-mL volumetric flasks. Samples were re-extracted three additional times, and the combined supernatants were brought to volume after rinsing the filter paper three times with 95% ethanol. Sample solutions were then transferred and stored in securely capped brown bottles.

Duplicate 300 µL aliquots from each extract were placed in 2-dram vials, and 100 µg of inositol (Sigma-Aldrich, St. Louis, MO, USA) was added as an internal standard to each sample. Samples were dried overnight in a Speed Vac. To remove contaminants, 250 mg of a MB-1 ion-exchange resin (UCW3600; generously provided by Purolite, Philadelphia, PA, USA) and a micro stir-bar were added to each sample. Deionized H_2O (1 mL) was added, and samples were stirred on a modular multistir plate (Cole-Parmer, Vernon Hills, IL) set at 680 rpm for 2 h. Samples were then centrifuged for 10 min at 3000 g_n, and the supernatant was decanted into a new vial. The supernatant was dried using a SpeedVac and placed in a desiccator overnight with lids loosened. Then, nitrogen and O-Bistrifluoroacetamide plus 1% trimethylsilyl (50 µL; Tokyo Chemical Industry, Tokyo, Japan) was added. Samples were vortexed for 30 s and incubated at room temp for 1 h. Next, dimethylformamide (100 µL) was added to the sample mixture, vortexed for 30 s, and incubated for another hour at room temperature before analysis. Samples appeared to be stable for at least 6 h after the addition of dimethylformamide. Multiple samples were prepared for morning injections, and new batches were prepared for afternoon injections onto a gas chromatograph.

Sugars were quantitated by injection onto a Varian 3400 gas chromatograph (Agilent Technologies, Santa Clara, CA, USA). Samples were vortexed for 30 s, and 0.5 µL was

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-16 via Open Access. This is an open access article distributed under the CC BY-NC license (https://creativecommons.org/licenses/by-nc/4.0/). https://creativecommons.org/licenses/by-nc/4.0/

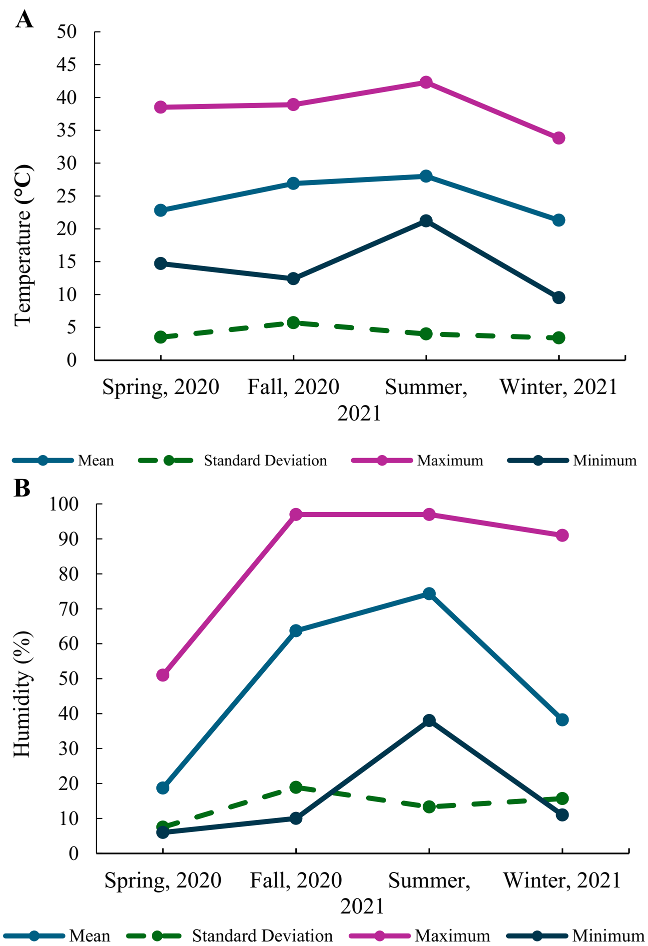

Fig. 1. (**A**) Temperature and (**B**) humidity in the greenhouse during the Spring 2020, Fall 2020, Summer 2021, and Winter 2021 growing seasons.

injected onto a DB-5 capillary column (column length, 30 m; diameter, 0.25 mm; film thickness, 0.25 μm; Agilent Technologies) equipped with a splitless injector held at 260 °C. The column temperature was initially held at 140 °C for 2 min, followed by a ramp of 20 °C/min until reaching 280 °C, and held for 9 min. Peaks were detected using a flame ionization detector held at 300 °C. Chromatographic data from the flame ionization detector signal was collected using Dionex Peak Net (Dionex Corporation, Sunnyvale, CA, USA) software. Sugars (glucose, fructose, and sucrose) were identified according to coelution with authentic standards and quantitated using inositol as the internal standard.

Sesquiterpene lactone extraction and analysis. The modified procedure of Ferioli and D'Antuono (2012) was used to extract SLs. Approximately 200 mg of ground freeze-dried plant material was accurately weighed in duplicate for free and bound SL determinations of each sample. Before extraction of the quadruplicate samples, 20 μg of santonin (Sigma-Aldrich, St. Louis, MO, USA) was added as the internal standard, followed by the addition of 3 mL of extraction solvent [MeOH, H_2O (4:1 v/v) +2% formic acid]. The samples were mixed for 15 s using a Vortex Genie stirrer (Scientific Industries, Bohemia, NY, USA) set at maximum speed and incubated at 60 °C for 30 min, with stirring every 10 min. After incubation, the samples were centrifuged at 3000 g_n for 20 min using a Speed Vac centrifuge. The supernatant was transferred into a separate vial, and extraction was repeated. Because of cloudiness, the combined supernatants were centrifuged again, decanted into a clean vial, and dried in a Speed Vac Centrifuge overnight (SVC-100H; Savant, Farmingdale, NY, USA).

The dried quadruplicate samples were reconstituted into deionized H_2O (3 mL) using vortex stirring for 20 s. To determine bound SLs, cellulase enzyme (*Aspergillus niger*, 25 mg, 1.1 units/mg; Sigma-Aldrich) was added to one duplicate set of the samples. Then, both free and bound duplicate sets of samples were vortexed and incubated at 40 °C for 2 h. Then, SLs were recovered into 2 mL of ethyl acetate by vortexing for 15 s, and samples were centrifuged at 3000 g_n in a Speed Vac centrifuge for 10 min to accommodate phase separation. The upper ethyl acetate phase was recovered, and the ethyl acetate SL recovery process was repeated twice. Combined ethyl acetate phases were evaporated to dryness for 3 h using a Speed Vac. The residues for both free and bound samples were dissolved again in 1 mL of methanol (>99%; EMD Millipore Corporation, Billerica, MA, USA) and overlaid with 5 mL of dichloromethane for further processing.

Both free and bound SL-containing fractions were processed by SPE according to Ferioli and D'Antuono (2012) using Extract-Clean silica columns (2.8 mL reservoir/500 mg silica sorbent; Alltech Associates Inc., Deerfield, IL, USA). The columns were preconditioned with 6 mL of dichloromethane/isopropanol (1:1 v/v) and equilibrated with 6 mL of dichloromethane. Samples were gravity-fed through the columns, and the eluate was dried for 2 h using a Speed Vac. Columns were reconditioned with 6 mL of dichloromethane/ethyl acetate (3:2 v/v), and the eluate was discarded.

Before high-performance liquid chromatography (HPLC) injection, samples were dissolved in 1 mL HPLC H_2O/MeOH (1:1 v/v), vortexed until the pellet was dissolved again, and filtered using a stainless-steel Millipore Filter apparatus (Millipore Corporation, Billerica, MA, USA) with a 0.45-μm nylon 66 filter (Supelco, Bellefonte, PA, USA) and Whatman 41 prefilter (Whatman International, Maidstone, England). The HPLC analyses were performed using a Thermo-Dionex Ultimate-3000 (ThermoFisher Scientific, Waltham, MA, USA) system with a gradient pump, autosampler, and PDA-1 diode array detector. Specifically, SLs were detected at 264 nm, and injection volumes were set for 10 μL. Separations were conducted using a Kinetex XB C18 column (5 μm; 250 × 4.6 mm) equipped with a C18 (4 × 3.0 mm) pre-column with cartridges placed in a Security Guard apparatus (Phenomenex, Torrance, CA, USA). Flow rate was set to 1.0 $mL \cdot min^{-1}$, and elution solvents were 10% and 55% acetonitrile in HPLC H_2O for solvent A and solvent B, respectively. The following eluent gradient program of 48 min was established: 100% solvent A for 5 min, followed by a linear gradient to 85% solvent B by 35 min, and then to 100% solvent B at 36 min. Solvent B was held at 100% for 8 min and then returned to 100% solvent A over 1 min. Initial conditions of 100% solvent A were held for 3 min before the next injection. Chromatograms were analyzed using a chromatography data system (Chromeleon 7; Thermo-Dionex, Waltham, MA, USA). The SLs were identified according to coelution with authentic standards (lactucin, 8-deoxylactucin, and lactucopicrin) and quantitated relative to santonin as an internal standard. Bound SLs were determined by subtracting bound SL samples (the cellulase-treated samples) from free SLs for lactucin, 8-deoxylactucin, and lactucopicrin.

Statistical analysis. Data were analyzed with SAS 9.4 (SAS Inc., Cary, NC, USA) using the PROC GLIMMIX procedure. The study included harvest season, cultivar, and the harvest season × cultivar interaction in four replications. When appropriate, differences among treatment means were determined using Tukey's least significant difference ($P \leq 0.05$).

Results

Effect of harvest season and lettuce cultivars on plant yield. The results showed that plant fresh weight was not significantly affected

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-16 via Open Access. This is an open access article distributed under the CC BY-NC license (https://creativecommons.org/licenses/by-nc/4.0/). https://creativecommons.org/licenses/by-nc/4.0/

Table 3. Effects of harvest season and cultivar on sesquiterpene lactone content, sugar content, and plant fresh weight of lettuce grown in nutrient flow technique hydroponics systems.

		Harvest season[i]	Cultivar[ii]	Harvest season × cultivar
Free SLs	Lactucin	***[iii]	**	*
	8-Deoxylactucin	***	***	***
	Lactucopicrin	***	***	**
Bound SLs	Lactucin	NS	NS	NS
	8-Deoxylactucin	*	***	NS
	Lactucopicrin	**	NS	NS
Total SLs	Total SLs	***	***	*
Soluble sugars	Fructose	***	*	NS
	Glucose	***	NS	NS
	Sucrose	***	***	*
Total sugar	Total sugar	***	NS	NS
	Sugar:SL ratio	***	***	**
	Plant fresh wt	***	***	NS

[i] Harvest seasons included Spring 2020, Fall 2020, early Summer 2021, and Winter 2021.
[ii] Twelve cultivars in this study were Jericho, Coastal Star, Parris Island, Buttercrunch, Nancy, Nevada, Cherokee, Sierra, Butter Red, Butter Green, Sweet Crisp Red, and Sweet Crisp Green.
[iii] NS, *, **, *** Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively.
SLs = sesquiterpene lactones.

by the harvest season × cultivar interaction. However, fresh weight was significantly different between seasons ($P < 0.001$) (Table 3), with the average plant fresh weight being greatest during both Spring 2020 and Fall 2020, followed by both Summer 2021 and Winter 2021 (Fig. 2A). Similarly, plant fresh weight varied significantly between cultivars ($P < 0.001$) (Table 3). The greatest plant fresh weight was found in romaine and Batavian market types, with romaine 'Jericho' showing the greatest plant fresh weight, and Salanova® 'Butter Red' and 'Butter Green' showing the lowest plant fresh weight (Fig. 2B). Notably, 'Summer Crisp Green' was the only Salanova® cultivar in the study that fell into the higher statistical grouping for plant weight.

Effect of harvest seasons, lettuce cultivars, and their interaction on lettuce quality. The results showed that there was a significant harvest season × cultivar interaction for all three free SLs (lactucin, 8-deoxylactucin, and lactucopicrin), total SLs, sucrose and sugar:SL ratio (Table 3). Furthermore, bound SLs, fructose, glucose, and total sugars were not significantly affected by the harvest season × cultivar interaction. Alternatively, free SLs, bound 8-deoxylactucin, fructose, sucrose, and sugar:SL ratio were significantly different for harvest seasons and cultivars. Bound lactucin was not significantly affected by either harvest season or cultivar. Bound lactucopicrin, glucose, and total sugars varied significantly between harvest seasons, but not between cultivars (Table 3).

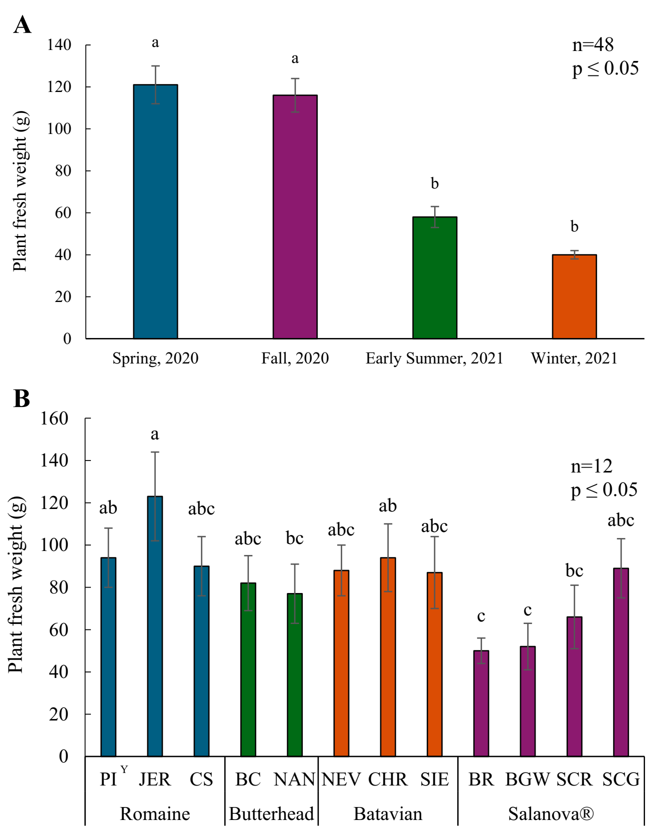

Fig. 2. Effects of (**A**) harvest season and (**B**) cultivar on plant fresh weight of lettuce. Lettuce cultivars: BC = Buttercrunch; BG = Butter Green; BR = Butter Red; CHR = Cherokee; CS = Coastal Star; JER = Jericho; NAN = Nancy; NEV = Nevada; PI = Parris Island; SCG = Sweet Crisp Green; SCR = Sweet Crisp Red; SIE = Sierra.

Effect of harvest season on bound SL and sugar concentrations. Significant main effects within harvest season were noted for bound SL concentrations (i.e., 8-deoxylactucin and lactucopicrin) and sugar concentrations (i.e., fructose, glucose, total sugars) (Table 4). In general, bound SL concentrations were almost a magnitude lower than their free counterparts (Tables 4 – 6). Specifically, bound 8-deoxylactucin was greatest in Spring 2020 and least in Fall 2020 and Winter 2021; bound lactucopicrin was greatest in Spring 2020, Summer 2021, and Fall 2020, and it was lowest in Winter 2021 (Table 4). However, fructose, glucose, and total sugar were greater in summer harvest seasons than in spring. Additionally, the fructose content was greatest in Fall 2020 and Summer 2021 and lowest in Spring 2020 and Winter 2021. Glucose was greatest in Summer 2021, followed by Fall 2021; both Spring 2020 and Winter 2021 showed similar concentrations. The total sugar content was greatest in Summer 2021 and lowest in Spring 2020.

Effect of lettuce type and cultivars on SL and sugar concentrations. Cultivar significantly affected the concentrations of bound 8-deoxylactucin and fructose (Table 5). Bound 8-deoxylactucin was greater for Batavian Cherokee than any other cultivar, including the other two Batavian types (i.e., Nevada and Sierra) (Table 5). Fructose concentrations were highest in two Batavian cultivars (Nevada and Sierra) and lowest in Salanova® Butter Red.

Effect of harvest seasons and lettuce cultivars on the free SL and sugar concentrations and sugar:SL ratio. Specifically, free lactucin was only different between cultivars in the summer season, with butterhead 'Buttercrunch' and Salanova® 'Butter Red' exhibiting the highest concentration (Fig. 2B). The concentration of free lactucopicrin predominated among the three SL in all cultivars except Batavian 'Cherokee', which had the greatest concentration of free 8-deoxylactucin among all cultivars and across all seasons. Additionally, total SLs appeared to be lower in the winter harvest across all cultivars. Predominantly, both Salanova® Butter cultivars appeared in the highest statistical groupings for total SLs in all seasons, whereas both Salanova® Sweet Crisp Green and Batavian Cherokee had the highest statistical grouping only once during the spring harvest. Alternatively, at least one cultivar within the romaine market type was in the lowest statistical grouping for total SLs in all seasons (i.e., Jericho in the spring and winter harvests, Parris Island in the summer and fall harvests, and Coastal Star in the summer harvest).

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-16 via Open Access. This is an open access article distributed under the CC BY-NC license (https://creativecommons.org/licenses/by-nc/4.0/). https://creativecommons.org/licenses/by-nc/4.0/

Table 4. Means, standard error, and mean separations for significant main effects within harvest season (Spring 2020, Fall 2020, early Summer 2021, Winter 2021) in nutrient flow technique hydroponic systems.

	SL concn ($\mu g \cdot g^{-1}$)		Sugar concn ($mg \cdot g^{-1}$)		
Harvest season	Bound 8-deoxylactucin	Bound lactucopicrin	Fructose	Glucose	Total sugar
Spring 2020[i]	3 ± 1 a[ii]	15 ± 2 a	25 ± 3 b	35 ± 3 c	67 ± 6 c
Fall 2020	2 ± 0 b	9 ± 4 ab	43 ± 5 a	57 ± 4 b	106 ± 9 b
Early Summer 2021	2 ± 1 ab	17 ± 2 a	45 ± 3 a	76 ± 2 a	149 ± 7 a
Winter 2021	1 ± 1 b	4 ± 1 b	21 ± 2 b	35 ± 1 c	92 ± 4 b

[i] Spring 2020 means do not include 'Butter Green' (n = 45).
[ii] Means (n = 48) within a column followed by the same lowercase letter are not significantly different according to the pairwise comparison in the model ($P \leq 0.05$).
SLs = sesquiterpene lactones.

Batavian cultivars exhibited total SLs in the lowest statistical grouping three times (Sierra in summer and fall and Nevada in summer). The butterhead and Salanova® cultivars were in the lowest statistical grouping in only one season (butterhead Buttercrunch in the fall harvest and Salanova® Sweet Crisp Green in the summer harvest). Also, cultivars only differed in the sucrose concentration in the summer harvest, with butterhead Nancy equivalent to Batavian Nevada and Sierra, all of which had a greater sucrose concentration than that of other cultivars. However, the sugar:SL ratio was only different among cultivars in the winter harvest, with romaine Jerricho exhibiting a greater ratio than that of butterhead Nancy, Batavian Cherokee, and all of the Salanova® cultivars.

Discussion

Within greenhouses, seasonal variations occur as a result of interactions between external weather patterns and how these patterns engage with the greenhouse's internal environment and existing passive heating and cooling systems (Santosh et al. 2017). These internal environmental fluctuations within a greenhouse affect plant growth, water uptake, overall plant health, and produce quality (Gruda 2005). Only a few other studies have established the seasonal influence on plant growth of hydroponically grown lettuce (Djidonou and Leskovar 2019; Fallovo et al. 2009). The current study found that head weight was greater in spring and fall production seasons than in the winter season (Fig. 2A). However, Djidonou and Leskovar (2019) observed greater head weight in hydroponically grown lettuce only in the spring season compared with that in the fall and winter growing seasons. The increased head weight was a product of a higher rate of leaf appearance under the favorable spring conditions of moderate temperature under optimal increasing light durations (Djidonou and Leskovar 2019). Alternatively, the current study with different cultivars observed a substantial decrease in head weight of lettuce harvested during summer and winter compared with that of lettuce harvested during spring and fall. The decrease in lettuce head weight in summer months could be attributable to the increasing hydroponic water temperature, which is a critical factor in hydroponic growth environments (Thakulla et al. 2021), caused by equilibrium of the uninsulated hydroponic table reservoir with the prevailing air temperature during the summer season. Additionally, lettuce is a long-day plant that produces undesirable flowers when exposed to light durations exceeding 12 h per day (i.e., summer months). Still, insufficient light durations in the winter can slow plant growth, leading to a low growth rate (Djidonou and Leskovar 2019). In the current study, longer light durations and higher water temperatures during summer months and shorter light durations and lower air and water temperatures during winter production seasons could have affected photosynthesis and respiration rates and ultimately resulted in reduced plant growth and yield.

Cultivar differences also have been noted in terms of plant fresh weight, highlighting the importance of cultivar selection to maximize plant growth, especially when growing in limited spaces such as greenhouses. Romaine market types were the greatest plant fresh weight group compared with other market types (Fig. 2B). Similarly, Afton et al. (2020) showed that romaine market types were heaviest compared with butterhead, crisphead, and loose leaf market types. Alternatively, the current study suggests that most romaine, butterhead, and Batavian lettuce heads were comparable in weight, with the exception of romaine 'Jericho', which yielded significantly more than butterhead 'Nancy' (Fig. 2B). Differences in weight between cultivars within a market type exist (Afton et al. 2020), indicating that while lettuce yield may be loosely categorized by market type, substantial differences in performance of individual cultivars must be considered when making production decisions for improved yield.

A significant cultivar × harvest season interaction in all free SLs, total SLs, and the sugar:SL ratio, but not for total sugars (Table 3), may indicate that any seasonal influence on lettuce flavor across cultivars depended more on SL concentrations than on total sugars for the lettuce market types and cultivars studied. According to Chadwick et al. (2016), a high sugar:SL ratio for lettuce correlated to less bitter perception by taste panelists and higher overall acceptance of the lettuce. Additionally, significant differences in the sugar:SL ratio occurred between cultivars in Summer 2021 and Winter 2021, with Winter 2021 exhibiting greater ratios (presumably less bitter flavor) than those of the Summer 2021 harvest (Table 4). The Winter 2021 increase in the sugar:SL ratio was related to total SL concentrations being approximately three-times lower while total sugar concentrations across cultivars were only less than two-times lower (Table 4). These data further support our findings that, compared to the sugar concentration, the SL concentration of lettuce may exert a larger influence on the sugar:SL ratio, which was not directly investigated during previous research. However, some caution may also be warranted when individual sugar concentrations are considered. Because plants from the Summer 2021 harvest season were significantly greater in fructose compared with those form the Winter 2021 harvest season, and because fructose is perceived to be twice as sweet as glucose (Chadwick et al. 2016; Gilbert et al. 2015), the summer lettuces may not have been perceived as bitter on the same scale as the winter lettuces despite exhibiting lower sugar:SL ratios (Table 4).

Table 5. Means, standard error, and mean separations for significant main effects within cultivar (four market types) in nutrient flow technique hydroponic systems.

Type	Cultivar	SL concn ($\mu g \cdot g^{-1}$) Bound 8-deoxylactucin	Sugar concn ($mg \cdot g^{-1}$) Fructose
Romaine	PI[i]	0 ± 0 b[ii]	37 ± 8 ab
	JER	0 ± 0 b	33 ± 7 ab
	CS	1 ± 1 b	37 ± 4 ab
Butterhead	BC	2 ± 1 b	34 ± 5 ab
	NAN	0 ± 0 b	31 ± 5 ab
Batavian	NEV	3 ± 1 b	46 ± 8 a
	CHR	12 ± 2 a	28 ± 5 ab
	SIE	2 ± 1 b	44 ± 9 a
Salanova®	BR	1 ± 1 b	17 ± 4 b
	BG[iii]	0 ± 0 b	34 ± 4 ab
	SCR	0 ± 0 b	35 ± 7 ab
	SCG	0 ± 0 b	28 ± 4 ab

[i] Lettuce cultivars: BC = Buttercrunch; BG = Butter Green; BR = Butter Red; CS = Coastal Star; JER = Jericho; NAN = Nancy; PI = Parris Island; SCG = Sweet Crisp Green; SCR = Sweet Crisp Red.
[ii] Means (n = 12) within a column followed by the same lowercase letter are not significantly different according to the pairwise comparison in the model ($P \leq 0.05$).
[iii] 'Butter Green' is missing Spring 2020 data.
SLs = sesquiterpene lactones.

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-16 via Open Access. This is an open access article distributed under the CC BY-NC license (https://creativecommons.org/licenses/by-nc/4.0/). https://creativecommons.org/licenses/by-nc/4.0/

Table 6. Lettuce quality means and standard error for interactions between cultivar and harvest seasons.

		SL concn (μg·g^{-1})				Sugar concn (mg·g^{-1})	
Type	Cultivar	Lactucin[i]	8-Deoxylactucin[i]	Lactucopicrin[i]	Total SLs	Sucrose	Sugar:SL
				Spring 2020			
R[ii]	PI[iii]	30 ± 12[iv]	0 ± 0 b[v]	72 ± 4 abc[iv]	115 ± 17 ab	4 ± 1	519 ± 119
	JER	9 ± 5	0 ± 0 b	31 ± 12 c	46 ± 17 b	5 ± 1	1219 ± 642
	CS	28 ± 1	5 ± 1 b	44 ± 10 bc	104 ± 20 ab	8 ± 2	1111 ± 242
BH	BC	17 ± 4	10 ± 3 b	51 ± 18 bc	96 ± 29 ab	13 ± 7	1497 ± 831
	NAN	17 ± 7	1 ± 0 b	112 ± 19 ab	124 ± 18 ab	11 ± 2	527 ± 212
BV	NEV	8 ± 0	9 ± 2 b	49 ± 9 bc	80 ± 12 ab	8 ± 2	1022 ± 306
	CHR	9 ± 2	78 ± 14 a	46 ± 4 bc	162 ± 16 a	5 ± 2	287 ± 82
	SIE	8 ± 1	11 ± 2 b	63 ± 5 bc	100 ± 3 ab	7 ± 3	762 ± 275
S	BR	18 ± 2	1 ± 0 b	105 ± 9 abc	154 ± 12 a	9 ± 5	582 ± 303
	BG	NA[v]	NA	NA	NA	NA	NA
	SCR	21 ± 10	0 ± 0 b	76 ± 48 abc	112 ± 32 ab	9 ± 3	742 ± 229
	SCG	23 ± 10	1 ± 0 b	131 ± 11 a	146 ± 21 a	6 ± 4	618 ± 265
				Fall 2020			
R	PI	5 ± 2	0 ± 0 c	41 ± 9 cd	50 ± 11 b	8 ± 5	2392 ± 969
	JER	21 ± 14	0 ± 0 c	31 ± 6 d	67 ± 31 ab	5 ± 1	2175 ± 1053
	CS	18 ± 5	2 ± 1 c	48 ± 8 bcd	79 ± 19 ab	6 ± 2	1905 ± 767
BH	BC	14 ± 5	2 ± 2 c	40 ± 8 d	62 ± 12 b	5 ± 2	1643 ± 273
	NAN	18 ± 9	1 ± 1 c	93 ± 19 abc	118 ± 24 ab	6 ± 1	1116 ± 262
BV	NEV	11 ± 6	4 ± 1 c	41 ± 4 cd	66 ± 11 ab	9 ± 2	2387 ± 884
	CHR	7 ± 2	46 ± 1 a	49 ± 7 bcd	123 ± 6 ab	5 ± 1	1050 ± 384
	SIE	6 ± 3	3 ± 0 c	37 ± 3 d	51 ± 4 b	9 ± 3	3202 ± 1290
S	BR	29 ± 8	0 ± 0 c	94 ± 15 ab	129 ± 23 ab	4 ± 0	479 ± 29
	BG	41 ± 29	12 ± 2 b	130 ± 20 a	194 ± 46 a	5 ± 1	520 ± 79
	SCR	25 ± 12	0 ± 0 c	48 ± 7 bcd	137 ± 66 ab	5 ± 1	941 ± 276
	SCG	24 ± 4	1 ± 1 c	103 ± 9 a	136 ± 8 ab	4 ± 1	675 ± 147
				Early Summer 2021			
R	PI	11 ± 0 ef	0 ± 0 c	93 ± 11 c	118 ± 11 c	16 ± 2 c	1091 ± 278 ab
	JER	36 ± 4 bc	1 ± 1 c	97 ± 11 bc	144 ± 11 bc	23 ± 3 bc	1052 ± 142 ab
	CS	13 ± 1 def	2 ± 1 c	76 ± 6 c	108 ± 7 c	23 ± 4 bc	1439 ± 381 ab
BH	BC	53 ± 5 a	14 ± 1 b	108 ± 9 abc	202 ± 25 abc	16 ± 1 c	576 ± 74 b
	NAN	17 ± 6 def	1 ± 1 c	106 ± 3 bc	167 ± 10 abc	53 ± 8 a	1068 ± 107 ab
BV	NEV	13 ± 3 def	7 ± 1 bc	74 ± 9 c	115 ± 8 c	40 ± 8 ab	2004 ± 339 ab
	CHR	7 ± 4 f	50 ± 8 a	58 ± 7 c	141 ± 20 bc	22 ± 0 bc	889 ± 79 b
	SIE	11 ± 2 ef	8 ± 1 bc	75 ± 5 c	113 ± 4 c	39 ± 5 abc	1519 ± 164 ab
S	BR	44 ± 7 ab	0 ± 0 c	174 ± 18 a	254 ± 31 a	16 ± 2 c	504 ± 120 b
	BG	27 ± 2 cd	1 ± 1 c	164 ± 34 ab	224 ± 44 ab	31 ± 5 bc	799 ± 229 b
	SCR	26 ± 3 cde	0 ± 0 c	99 ± 11 bc	134 ± 13 bc	29 ± 5 bc	1190 ± 147 ab
	SCG	20 ± 2 cdef	0 ± 0 c	86 ± 8 c	124 ± 13 c	28 ± 2 bc	1194 ± 60 ab
				Winter 2021			
R	PI	3 ± 1	1 ± 1 b	20 ± 4 abc	25 ± 5 bc	36 ± 2	4357 ± 1049 ab
	JER	2 ± 1	0 ± 0 b	7 ± 1 c	12 ± 1 c	43 ± 3	8966 ± 517 a
	CS	4 ± 1	0 ± 0 b	11 ± 1 bc	22 ± 3 bc	34 ± 3	5696 ± 854 ab
BH	BC	2 ± 1	0 ± 0 b	10 ± 1 c	18 ± 1 bc	33 ± 6	5239 ± 1345 ab
	NAN	10 ± 6	1 ± 1 b	29 ± 2 ab	44 ± 9 ab	39 ± 9	1858 ± 564 b
BV	NEV	2 ± 1	1 ± 0 b	14 ± 3 ab	28 ± 8 bc	43 ± 4	4991 ± 1131 ab
	CHR	12 ± 4	11 ± 0 a	9 ± 1 c	44 ± 3 ab	36 ± 6	2986 ± 556 b
	SIE	3 ± 1	2 ± 2 b	16 ± 3 abc	24 ± 4 bc	46 ± 6	5088 ± 1474 ab
S	BR	10 ± 4	0 ± 0 b	24 ± 2 abc	45 ± 7 ab	30 ± 7	1431 ± 202 b
	BG	22 ± 11	1 ± 1 b	30 ± 9 a	61 ± 7a	38 ± 5	1356 ± 124 b
	SCR	16 ± 11	1 ± 1 b	14 ± 3 bc	37 ± 9 abc	37 ± 7	3261 ± 970 b
	SCG	5 ± 1	0 ± 0 b	21 ± 6 abc	29 ± 7 bc	28 ± 5	3989 ± 1598 b

[i] All individual SLs were free, and the total SLs comprised the cumulative total of all free and bound SLs.
[ii] Lettuce market types: R = Romaine; BH = Butterhead; BV = Batavian; S = Salanova®.
[iii] Lettuce cultivars: BC = Buttercrunch; BG = Butter Green; BR = Butter Red; CS = Coastal Star; JER = Jericho; NAN = Nancy; PI = Parris Island; SCG = Sweet Crisp Green; SCR = Sweet Crisp Red.
[iv] Values without an adjacent lowercase letter indicate nonsignificant effects for the variable. Means (n = 5) within a column separated by different letters are significantly different at $P \leq 0.05$.
[v] NA = not analyzed.
SLs = sesquiterpene lactones.

Romaine and Batavian cultivars (with the exception of Cherokee) consistently performed in lower statistical groupings for the concentration of the most abundant SLs found in the lettuce (i.e., lactucopicrin) (Table 5). Because lactucopicrin predominated in concentration among the other SLs evaluated in this study, with the exception of 'Cherokee' (in which 8-deoxylactucin predominated), a lower abundance of lactucopicrin among cultivars may decrease the assumed comparative perception of bitterness. Hence, romaine and Batavian (except 'Cherokee') market types may have less assumed bitterness than that of other cultivars in the study. Likewise, Seo et al. (2009) used that bitterness score and concluded that lactucopicrin exerted the greatest influence on lettuce bitter off-flavor in Korean lettuces, in which lactucopicrin was also a major SL. The significant harvest season × cultivar interactions for free lactucopicrin and

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-16 via Open Access. This is an open access article distributed under the CC BY-NC license (https://creativecommons.org/licenses/by-nc/4.0/). https://creativecommons.org/licenses/by-nc/4.0/

total SL suggest that environmental factors such as temperature and daylength can affect SL production in lettuce (Hao et al. 2018).

Seasonal effects influenced SLs in most cultivars in this study that appear to be affected by increased stress associated with the summer heat and longer daylengths (Table 6), with the exception of Butter Red and Butter Green in the Salanova® market class. Salanova® market types recorded the highest total SL groupings compared with those of most cultivars, regardless of the production season. However, very little is known about putative flavor characteristics for the Salanova® lettuce market class. Specifically, substantially higher free and total SLs and corresponding lower sugar:SL ratios were recorded for the Butter Red and Butter Green Salanova® cultivars compared with those of other market types (Table 6), which might indicate a greater tendency toward bitter flavor perception for these cultivars. Alternatively, the Summer Crisp Salanova® cultivars exhibited numerically higher (but not significantly) sugar:SL ratios compared with those of the Butter cultivars, perhaps indicating that certain cultivars within this market type may have differences in bitter flavor perception.

Conclusion

Cultivar selection within market type is critical because cultivar performance and taste vary within different cultivars in a market type with varying seasons. Generally, lettuces with lower SL concentrations and/or higher sugar concentration have less perceived bitterness, and a greater sugar:SL ratio is associated with increased lettuce palatability because sweetness from sugars can mask bitterness from SL. Hence, romaine and Batavian market types (excluding ‘Cherokee’) were considered to be the best performers in the summer in a greenhouse setting, with the greatest plant fresh weight, lowest SL concentration, and greater sugar:SL ratio; butterhead cultivars performed similarly in spring. The differences in putative flavor were less pronounced between different cultivars in winter because of lower concentrations of SL and sugars. The current study will help growers select cultivars that yield higher plant fresh weight and have a better perception of flavor for different production seasons in greenhouse environments. Because of greater plant fresh weight, low SL concentrations, and greater sugar:SL ratio, we recommend romaine and Batavian market types (except ‘Cherokee’) during the summer months in hydroponic crop culture. In addition to cultivars, sugar and SL concentrations are affected by environmental conditions. Hence, future research should focus on strategies to mitigate bitterness compounds production and increase accumulation of sugars in different lettuce market types and cultivars.

References Cited

Afton WD, Fontenot KK, Kuehny JS, Motsenbocker CE. 2020. Evaluation of yield, marketability, and nitrate levels of lettuce cultivars produced in southern Louisiana. HortTechnology. 30(5): 632–637. https://doi.org/10.21273/HORTTECH04642-20.

Calín-Sánchez Á, Lipan L, Cano-Lamadrid M, Kharaghani A, Masztalerz K, Carbonell-Barrachina ÁA, Figiel A. 2020. Comparison of traditional and novel drying techniques and its effect on quality of fruits, vegetables and aromatic herbs. Foods. 9(9):1261. https://doi.org/10.3390/foods9091261.

Chadwick M, Gawthrop F, Michelmore RW, Wagstaff C, Methven L. 2016. Perception of bitterness, sweetness and liking of different genotypes of lettuce. Food Chem. 197(Pt A):66–74. https://doi.org/10.1016/j.foodchem.2015.10.105.

Davies H. 1988. Rapid determination of glucose, fructose, and sucrose in potato tubers cy capillary gas chromatography. Potato Res. 31:569–572. https://doi.org/10.1007/BF02361846.

Djidonou D, Leskovar DI. 2019. Seasonal changes in growth, nitrogen nutrition, and yield of hydroponic lettuce. HortScience. 54(1):76–85. https://doi.org/10.21273/HORTSCI13567-18.

Fallovo C, Rouphael Y, Rea E, Battistelli A, Colla G. 2009. Nutrient solution concentration and growing season affect yield and quality of *Lactuca sativa* L. var. acephala in floating raft culture. J Sci Food Agric. 89(10):1682–1689. https://doi.org/10.1002/jsfa.3641.

Ferioli F, D’Antuono LF. 2012. An update procedure for an effective and simultaneous extraction of sesquiterpene lactones and phenolics from chicory. Food Chem. 135:243–250. https://doi.org/10.1016/J.FOODCHEM.2012.04.079.

Gargaro M, Murphy RJ, Harris ZM. 2023. Let-us investigate: A meta-analysis of influencing factors on lettuce crop yields within controlled-environment agriculture systems. Plants. 12(14):2623. https://doi.org/10.3390/plants12142623.

Gilbert JL, Guthart MJ, Gezan SA, de Carvalho MP, Schwieterman ML, Colquhoun TA, Bartoshuk LM, Sims CA, Clark DG, Olmstead JW. 2015. Identifying breeding priorities for blueberry flavor using biochemical, sensory, and genotype by environment analyses. PLoS One. 10(9):e0138494. https://doi.org/10.1371/journal.pone.0138494.

Gruda N. 2005. Impact of environmental factors on product quality of greenhouse vegetables for fresh consumption. Crit Rev Plant Sci. 24(3):227–247. https://doi.org/10.1080/07352680591008628.

Hao J-H, Zhang L-L, Li P-P, Sun Y-C, Li J-K, Qin X-X, Wang L, Qi Z-Y, Xiao S, Han Y-Y, Liu C-J, Fan S-X. 2018. Quantitative proteomics analysis of lettuce (*Lactuca sativa* L.) reveals molecular basis-associated auxin and photosynthesis with bolting induced by high temperature. Int J Mol Sci. 19(10):2967. https://doi.org/10.3390/ijms19102967.

Hernandez E, Timmons MB, Mattson NS. 2020. Quality, yield, and biomass efficacy of several hydroponic lettuce (*Lactuca sativa* L.) cultivars in response to high pressure sodium lights or light emitting diodes for greenhouse supplemental lighting. Horticulturae. 6(1):7. https://doi.org/10.3390/horticulturae6010007.

Holmes SC, Wells DE, Pickens JM, Kemble JM. 2019. Selection of heat tolerant lettuce (*Lactuca sativa* L.) cultivars grown in deep water culture and their marketability. Horticulturae. 5(3):50. https://doi.org/10.3390/horticulturae5030050.

Khan M. 2018. Effect of high temperature and exposure duration on stem elongation of iceberg lettuce. Pak J Agric Sci. 55. https://doi.org/10.21162/PAKJAS/18.655.

Lee O, Sugiyama N. 2006. Changes in carbohydrate composition in lettuce flower stalks during development. J Hortic Sci Biotechnol. 81(5): 928–932. https://doi.org/10.1080/14620316.2006.11512161.

Lei C, Engeseth NJ. 2021. Comparison of growth characteristics, functional qualities, and texture of hydroponically grown and soil-grown lettuce. LWT. 150:111931. https://doi.org/10.1016/j.lwt.2021.111931.

Li J, Abbas K, Wang L, Gong B, Hou S, Wang W, Dai B, Xia H, Wu X, Lü G, Gao H. 2023. Drought resistance index screening and evaluation of lettuce under water deficit conditions on the basis of morphological and physiological differences. Front Plant Sci. 14:1228084. https://doi.org/10.3389/fpls.2023.1228084.

Maness N. 2010. Extraction and analysis of soluble carbohydrates, p 341–370. In: Sunkar R (ed). Plant stress tolerance: methods and protocols. [accessed 1 Apr 2022].

Šalagovič J, Vanhees D, Verboven P, Holsteens K, Verlinden B, Huysmans M, Van de Poel B, Nicolaï B. 2024. Microclimate monitoring in commercial tomato (*Solanum Lycopersicum* L.) greenhouse production and its effect on plant growth, yield and fruit quality. Front Hortic. https://doi.org/10.3389/fhort.2024.1425285.

Santosh DTT, Singh VK, Reddy ARG. 2017. Micro climate control in greenhouse. IntJCurr MicrobiolAppSci. 6(3):1730–1742. https://doi.org/10.20546/ijcmas.2017.603.199.

Seo MW, Yang DS, Kays SJ, Lee GP, Park KW. 2009. Sesquiterpene lactones and bitterness in korean leaf lettuce cultivars. HortScience. 44(2):246–249. https://doi.org/10.21273/HORTSCI.44.2.246.

Sublett WL, Barickman TC, Sams CE. 2018. The effect of environment and nutrients on hydroponic lettuce yield, quality, and phytonutrients. Horticulturae. 4(4):48. https://doi.org/10.3390/horticulturae4040048.

Thakulla D, Dunn B, Hu B, Goad C, Maness N. 2021. Nutrient solution temperature affects growth and °Brix parameters of seventeen lettuce cultivars grown in an NFT hydroponic system. Horticulturae. 7(9):321. https://doi.org/10.3390/horticulturae7090321.

US Department of Agriculture. 2023. U.S. lettuce production shifts regionally by season. http://www.ers.usda.gov/data-products/chart-gallery/gallery/chart-detail/?chartId=106516. [accessed 15 Oct 2024].

Wang L, Ning S, Zheng W, Guo J, Li Y, Li Y, Chen X, Ben-Gal A, Wei X. 2023. Performance analysis of two typical greenhouse lettuce production systems: Commercial hydroponic production and traditional soil cultivation. Front Plant Sci. 14:1165856. https://doi.org/10.3389/fpls.2023.1165856.

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-16 via Open Access. This is an open access article distributed under the CC BY-NC license (https://creativecommons.org/licenses/by-nc/4.0/). https://creativecommons.org/licenses/by-nc/4.0/