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DIFFERENT NITRATE/AMMONIUM RELATIONSHIPS AND LIGHT INTENSITY AND THEIR EFFECT ON GROWTH AND NUTRITIONAL CONTENT OF HYDROPONIC LETTUCE (*Lactuca sativa* L.)

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ABSTRACT

Lettuce is the most consumed leafy crop worldwide and the high levels of nitrates in its leaves can be carcinogenic. The objective of this study was to evaluate the effect of different nitrate/ammonium ratios in the nutrient solution and two light intensities (200 and 400 μmol·m⁻²·s⁻¹) on the growth and nutrient concentration of lettuce plants under hydroponic conditions. Controlled environment chambers (light and temperature) and a floating root system were used with the application of Steiner's nutrient solution modified for each of the treatments. Treatments consisted of combinations of three nitrate/ammonium ratios (100/0, 75/25, and 50/50) and two LED light intensities. The variables evaluated were root volume, fresh and dry weight of the leaves, dry weight of the root, total dry weight and the mineral composition of the leaves. The experimental design was completely randomized with a 3 × 2 factorial arrangement with six repetitions. For the statistical analysis of the data, an analysis of variance and a comparison of means test were performed (Tukey, $p≤0.05$). The results showed that increasing the proportion of ammonium reduced the volume and dry weight of the root without affecting the fresh weight of the leaf. Increasing the light intensity increased the root volume and the fresh and dry weight of the leaves, while ammonium reduced the levels of potassium, calcium and nitrates in the leaves, especially when an intensity of 400 μmol·m⁻²·s⁻¹ was applied. These results suggest that optimizing nitrate/ammonium ratios and light conditions improves biomass production and nutritional quality of hydroponic lettuce.
Additional Keywords: Artificial light, controlled environment agriculture, nitrate reduction, nutrient solution, plant nutrition

RESUMEN

Diferentes relaciones nitrato/amonio e intensidad luminosa sobre el crecimiento y contenido nutricional de lechuga (*Lactuca sativa* L.) en cultivo hidropónico

La lechuga es el cultivo de hoja más consumido mundialmente y los altos niveles de nitratos en sus hojas pueden ser carcinogénicos. El objetivo de este estudio fue evaluar el efecto de distintas proporciones nitrato/amonio en la solución nutritiva y dos intensidades de luz (200 y 400 μmol·m⁻²·s⁻¹) en el crecimiento y concentración nutrimental de plantas de lechuga bajo condiciones de hidroponía. Se usaron cámaras de ambiente controlado (luz y temperatura) y un sistema de raíces flotantes con la aplicación de la solución nutritiva de Steiner modificada para cada uno de los tratamientos. Los tratamientos consistieron en combinaciones de tres proporciones de nitrato/amonio (100/0, 75/25, y 50/50) y dos intensidades de luz LED. Las variables evaluadas fueron el volumen radical, peso fresco y seco de las hojas, peso seco de la raíz, peso seco total y la composición mineral de las hojas. El diseño experimental fue un completamente al azar con un arreglo factorial 3×2 con seis repeticiones. Para el análisis estadístico de los datos se realizó un análisis de varianza y una prueba de comparación de medias (Tukey, $p≤0,05$). Los resultados mostraron que al aumentar la proporción de amonio se redujo el volumen y peso seco de la raíz sin afectar el peso fresco de la hoja. Al aumentar la intensidad de luz se incrementó el volumen radical, el peso fresco y seco de las hojas, mientras que el amonio redujo los niveles de potasio, calcio y nitratos en las hojas, especialmente cuando se aplicó una intensidad de 400 μmol·m⁻²·s⁻¹. Estos resultados sugieren que al optimizar las proporciones de nitrato/amonio y las condiciones de luz, mejora la producción de biomasa y la calidad nutricional de la lechuga hidropónica.
Palabras clave adicionales: Agricultura en ambiente controlado, luz artificial, nutrición vegetal, reducción de nitratos, solución nutritiva

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INTRODUCTION

Lettuce (*Lactuca sativa* L.) is a crop of great economic importance at a national and international level due to its high demand in the market, with a national and global production in

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2023 of 552940.26 and 28084971.27 t (FAOSTAT 2025) respectively.

It is consumed fresh in salads and as a side dish, being low in calories and recommended in diets (Baslam *et al*., 2013). Moreover, the soilless cultivation system is an alternative to obtaining vegetables of high hygienic quality, with higher yield and value than soil cultivation (Capera *et al*., 2017). In hydroponic lettuce production using a floating root system without substrates, plants develop directly in a nutrient solution, achieving greater efficiency in water supply and mineral nutrition, while avoiding climatic and light limitations (Moreno *et al*., 2015; Saavedra, 2017). This method also results in less presence of diseases and pests (Raviv and Lieth, 2008).

Regarding crop nutrition, nitrogen is one of the elements that influences the growth and development of plants (Chowdhury and Das, 2015). Absorbed nitrogen accounts for 1.5 to 5 % of the total dry weight and is the only nutrient that can be absorbed in the form of anion (NO_3^-), cation (NH_4^+), and amino acids (Näsholm *et al*., 2009). Fifty percent of the total nitrogen is part of high-molecular-weight compounds, such as proteins and nucleic acids, while the other 50 % is present as inorganic compounds, nitrates, and ammonium (Azcon and Talon, 2000). Lettuce plants grow best when they have access to a combination of both nitrogenous forms, according to Lara *et al.* (2019). However, the use of ammonium (NH_4^+) is restricted in soilless crops because at high concentrations it can be toxic to plants (Cramer and Lewis, 1993). Some studies report proportions of 75 % to 25 % NO_3^-/NH_4^+ to mitigate ammonium toxicity. Furthermore, Lu *et al*. (2009) stated in their research that tomato plants exhibit greater root volume and aerial growth when a proportion of 75 % NO_3^- and 25 % NH_4^+ in the nutrient solution is applied.

In contrast, using nitrate alone in the nutrient solution as a nitrogen source can cause NO_3^- accumulation in plant tissue. Nitrates are reduced to nitrites and N-nitrous compounds, which can be carcinogenic for people who consume raw lettuce (Karwowska and Kononiuk, 2020); about 80 % of nitrates in the typical diet are incorporated through leafy vegetables (ASTRE, 2015). When nitrate absorption exceeds assimilation, nitrates (NO_3^-) can accumulate in the vacuoles of cells (Masclaux *et al*., 2010).

It is known that plants have sophisticated mechanisms for selecting light energy, which is necessary for photosynthesis (Nguy *et al*., 2015). Therefore, the quantity and quality of light are determining factors for plant growth and development (Casierra and Peña, 2015). Specifically, plant growth can be affected by the quality, intensity, and duration of light (Jiao *et al*., 2007). Most plants can respond in different ways to light quality (Fukuda *et al*., 2008). The magnitude of this effect depends in part on the plant species and its genetic variants, as well as factors such as light quality and photoperiod (Chen *et al*., 2014).

Studies on lettuce cultivation show that increasing light intensity promotes growth and weight accumulation due to increased photosynthetic activity and mineral absorption (Hunter and Burritt, 2004; Chen *et al*., 2014). The magnitude of this effect depends partly on the plant species and its variety, as well as factors such as light quality (Enciso *et al*., 2024) and photoperiod (Chen *et al*., 2014). Light and photoperiod play a role in controlling nitrate content, which is a key aspect related to the quality of leafy vegetables (Cavaiuolo and Ferrante, 2014). It has been proposed that continuous light reduces the NO_3^- content in lettuce, stimulating the activity of the nitrate reductase enzyme, which facilitates NO_3^- assimilation and metabolism (Samuolienė *et al*., 2011). The productivity and quality of lettuce can be positively affected by modulating light quality and intensity, as well as other cultural practices (Loconsole *et al*., 2019). The concept of quality involves factors related to physical, chemical, nutritional, and biological aspects that producers must consider when competing in the market (Chiesa, 2010). Due to the above, the European Commission has legislated indicating the maximum nitrate contents allowed for lettuce grown in greenhouses ranging from 2500 to 4500 $mg \cdot kg^{-1}$ of NO_3 (Carrasco *et al*., 2006). The Joint Expert Committee of the Food and Agriculture (JECFA) Organization of the United Nations/World Health Organization and the European Commission (EC) Scientific Committee on Food have also set an acceptable daily intake for nitrate of 0-3.7 $mg \cdot kg^{-1}$ body weight (Santamaria, 2006).

The use of different intensities of artificial lighting using light-emitting diodes (LEDs) and the addition of ammonium to the nutrient solution can become important factors in reducing nitrate concentration in hydroponic lettuce. Therefore, the objective of this research was to evaluate the effect of different nitrate/ammonium ratios on the nutrient solution, in combination with different light intensities on the growth and nitrate content in hydroponic lettuce leaves, as well as the growth and weight of roots.

MATERIALS AND METHODS

Study area location. The present research was carried out in the Laboratory of Plant Physiology and Anatomy of the Agronomy Faculty of the Autonomous University of Sinaloa, México, 24°37'29.8” N latitude and 107°26'37.7” W longitude.

Establishment of the crop

Genetic material, experimental conditions and transplanting. Lettuce plants of the 41-co1225 (Rijk Zwaan) variety were used for this study. The experiment was conducted in two growth chambers, each with 1.94 m^3 of usable space (1.66 x 1.62 x 0.72 m), both maintained under controlled conditions with temperatures ranging from 22 to 24 °C, relative humidity between 60 and 80 %, and ambient carbon dioxide concentration.

Sowing was performed in 200-cavity polystyrene trays filled with peat (PRO-MIX® FLX, Premier Tech Horticulture, USA). Once the plants developed their first five true leaves (30 days after sowing), they were transplanted into containers filled with nutrient solution. Three containers (50 x 34 x 21 cm), each holding 20 liters of nutrient solution, were placed in each growth chamber. Six lettuce plants were positioned on the surface of each container.

Lighting was provided by transparent white LED tubes (T8 LED 30 W, 6500 K, 2400 lm; MRGL-08, Megaluz, Mexico). The first growth chamber contained 9 white LED tubes to achieve a total light intensity of 400 $\mu mol \cdot m^{-2} \cdot s^{-1}$, while the second growth chamber had 5 white LED tubes, resulting in a total light intensity of 200 $\mu mol \cdot m^{-2} \cdot s^{-1}$. Transplanting was carried out on May 13, 2021.

Hydroponic management in the floating root system and preparation of nutrient solutions. Plants were placed in floating root baskets, which were positioned over perforations in the lids of the containers, ensuring that the roots were fully immersed in the nutrient solution. The nutrient solutions were prepared using water-soluble inorganic salts dissolved in distilled water. Modifications were made to the universal nutrient solution (Steiner, 1984) to accommodate the different nitrate/ammonium ratios, as shown in Table 1. To these solutions, microelements were added from the Quelato Fullmix B source, which has the following composition: iron (Fe) 3.00 $mg \cdot L^{-1}$, manganese (Mn) 1.48 $mg \cdot L^{-1}$, boron (B) 0.28 $mg \cdot L^{-1}$, copper (Cu) 0.12 $mg \cdot L^{-1}$, zinc (Zn) 0.24 $mg \cdot L^{-1}$ and molybdenum (Mo) 0.08 mg L^{-1}.

Irrigation. The plant roots were continuously immersed in the nutrient solution, which was completely renewed every seven days. The pH of the solution was maintained between 5.5 and 6.0 by adjusting with sulfuric acid (H_2SO_4) and monitored using a pH/EC/TDS meter (HI-98130, Hanna) and an electrical conductivity (EC) of 2.0 $dS \cdot m^{-1}$. To ensure adequate oxygenation, oxygen pumps were employed to maintain dissolved oxygen levels in the nutrient solution at 6 to 8 $mg \cdot L^{-1}$.

Experimental design and treatments. A completely randomized design was used in a 3 x 2 factorial treatment arrangement, composed of six treatments and six repetitions. Factor A represented the nitrate/ammonium ratio with three levels: 100/0, 75/25 and 50/50. The B factor corresponded to the light intensity with two levels: 200 and 400 $\mu mol \cdot m^{-2} \cdot s^{-1}$.

The photosynthetic photon flux density (PPFD, in $\mu mol \cdot m^{-2} \cdot s^{-1}$) was measured using a ceptometer (AccuPAR LP-80, Decagon). Each experimental unit consisted of a single lettuce plant, resulting in a total of six treatments, each with six replications, for a total of 36 experimental units.

Since the Steiner solution did not include NH_4^+ as a nitrogen source, the modification involved adding a certain concentration of NH_4^+, which reduced the equivalent amount of NO_3^- to a total concentration of 12 $meq \cdot L^{-1}$. As the amount of NO_3^- decreased, the relative concentration of the anions (NO_3^-, $H_2PO_4^-$ and SO_4^{-2}) also decreased. These anions must maintain a total concentration of 20 meq·L^{-1}, so the concentrations of $H_2PO_4^-$ and SO_4^{-2} were increased until they matched the concentration of NH_4^+, maintaining the mutual ratio of $H_2PO_4^-$ and SO_4^{-2}. The addition of NH_4^+ to the nutrient solution increased the concentration of the cations (K^+, Ca^{+2}, Mg^{+2} and NH_4^+). Consequently, the amount of NH_4^+ added was subtracted from the concentrations of K^+, Ca^{+2} and Mg^{+2}, allowing the total concentration of the cations to remain at 20 meq·L^{-1}, always maintaining the mutual relationship between K^+, Ca^{+2}, and Mg^{+2}. The ion concentrations were adjusted using a correction factor (0.024) proposed by Steiner (1984), because the modifications changed the osmotic pressure of the nutrient solution. The solutions had an osmotic pressure of -0.072 MPa and an electrical conductivity (EC) of 2.0 dS·m^{-1}.

Table 1. Chemical composition of nutrient solutions and light intensities used in each treatment of the experiment

Treatment (NO_3/NH_4 x PPFD)	PPFD (µmol·m^{-2} s^{-1})	NO_3^-	$H_2PO_4^-$	SO_4^{2-}	K^+	Ca^{2+}	Mg^{2+}	NH_4^+
		mol$_c$ ·m^{-3}						
100/0 x 200	200	12.00	1.00	7.00	7.00	9.00	4.00	0.00
75/25 x 200	200	9.00	1.37	9.62	5.95	7.65	3.40	3.00
50/50 x 200	200	6.00	1.75	12.25	4.90	6.30	2.80	6.00
100/0 x 400	400	12.00	1.00	7.00	7.00	9.00	4.00	0.00
75/25 x 400	400	9.00	1.37	9.62	5.95	7.65	3.40	3.00
50/50 x 400	400	6.00	1.75	12.25	4.90	6.30	2.80	6.00

Radical volume (cm^3), root dry weight (g), fresh and dry leaf weight and total dry weight (g). The lettuce plants were harvested 29 days after transplanting, at the stage of commercial maturity. The fresh weight of each lettuce was measured using a precision balance (CP622, Sartorius, Germany). Root volume was determined using the water displacement method (Böhm, 1979). Additionally, the dry weights (70 °C, 72 h) of leaves and roots were recorded separately on an analytical balance.

P, K, Ca, Mg, NO_3 and NH_4 contents in leaves. The mineral composition of lettuce leaves was evaluated 29 days after the initiation of treatments. The leaves were dried in an oven (9053L, Ecoshel, USA) at 70 °C for 72 hours. Subsequently, they were processed in an electric mill (MOGRA1, Surtek, Mexico) and passed through a 40-mesh sieve. The resulting material underwent dry digestion in a muffle furnace (DTT 434, Caisa, Mexico) at 550 °C for 5 hours. Two milliliters of hydrochloric acid were added to the residue, and the mixture was heated until the liquid evaporated. Distilled water was then added, and the solution was filtered through filter paper. The filtrate was diluted to 100 mL in a volumetric flask.

These filtrates were used to determine the concentrations of phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg) using methods proposed by Motsara and Roy (2008). Phosphorus was measured by colorimetry using the molybdophosphoric complex with ascorbic acid (AOAC, 1980). Potassium, calcium, and magnesium were measured by atomic absorption spectrometry (Aanalyst 200, Perkin Elmer, USA), following the methodology of Rodríguez and Rodríguez (2015).

Nitrate (NO_3) content was determined using the Cataldo method, and reduced nitrogen (NH_4) content was measured by the Nessler method, as described by Alcántar and Sandoval (1999). In the Nessler method, 0.2 g of dry plant material was weighed, and 10 mL of phosphate buffer was added. The mixture was shaken for 1 hour in a vortex at intervals of 10 to 15 minutes and then filtered into glass bottles. Two milliliters of Nessler's reagent were added to the filtrate, which was then shaken and allowed to rest for 10 minutes. The samples were read at 420 nm, and the concentrations of ammonium (NH_4) in the plant tissue were calculated by interpolating the absorbance readings using the factor obtained from the slope of the calibration curve, considering the dilutions and weight of the plant material.

Statistical Analysis. An analysis of variance (ANOVA) was conducted on the data obtained from the studied variables. The primary factors evaluated in the factorial design were the nitrate/ammonium ratio in the nutrient solution (100/0, 75/25, and 50/50) and light intensity (200 and 400 µmol·m⁻²·s⁻¹), as well as their interaction. Mean comparisons were performed using the Tukey test ($p \leq 0.05$). The statistical analyses were performed with the Statistica program (StatSoft, 2004).

RESULTS AND DISCUSSION

Radical volume and root dry weight. The results of radical volume and root dry weight of lettuce are presented in Figure 1. Both variables were significantly ($p \leq 0.05$) affected by the nitrate/ammonium ratio and photosynthetic photon flux density (PPFD). The 50/50 nitrate/ammonium ratio with 200 $\mu mol \cdot m^{-2} \cdot s^{-1}$ PPFD decreased radical volume by 28 % (Figure 1A) and root dry weight by 9 % (Figure 1B) compared to plants without NH_4 application (100/0).

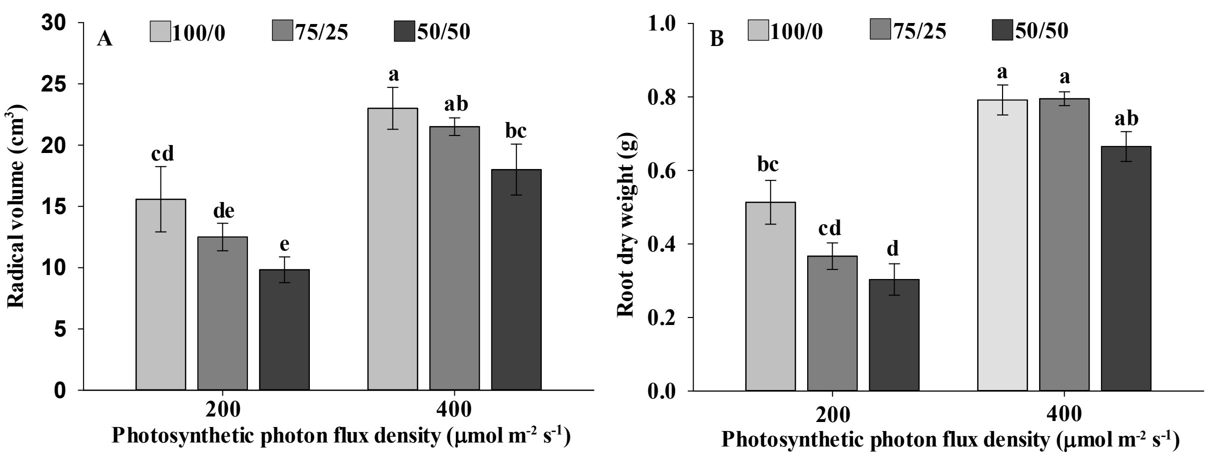

Figure 1. Effect of the interaction between the nitrate/ammonium ratio in nutrient solution (100/0, 75/25, and 50/50) and photosynthetic photon flux density (200 and 400 $\mu mol \cdot m^{-2} \cdot s^{-1}$) on the radical volume (A) and root dry weight (B) of lettuce plants. Means ± standard error with identical letters above the bars indicate no significant difference. (Tukey, $p \leq 0.05$).

These reductions in radical volume and root dry weight can be attributed to ammonium-induced medium acidification, which cannot be stored in the root or quickly converted into organic compounds, leading to toxicity (Magalhaes and Huber, 1989; Song, *et al.*, 2022). This is consistent with what was reported by Parra *et al.* (2012), who observed a considerable decrease in radical volume in tomato plants when the nitrate/ammonium ratio was increased to 70/30, suggesting high sensitivity to the nitrate/ammonium ratio. Regarding root dry weight, this decrease is related to Lara *et al.* (2019), who reported that a 50/50 nitrate/ammonium ratio caused 16 % less root dry weight in lettuce compared to other treatments.

In contrast, a PPFD of 400 $\mu mol \cdot m^{-2} \cdot s^{-1}$ led to a 65 % increase in radical volume (Figure 1A) and a 90 % increase in root dry weight (Figure 1B) compared to a PPFD of 200 $\mu mol \cdot m^{-2} \cdot s^{-1}$. These findings align with those of Weiguo *et al.* (2012) and Avendaño *et al.* (2020), who reported that lettuce plants cultivated under light intensities ranging from 400 to 600 $\mu mol \cdot m^{-2} \cdot s^{-1}$ exhibited higher quality and growth.

Fresh and dry leaf weight and total dry weight. The nitrate/ammonium ratio did not have a statistically significant effect ($p > 0.05$) on the fresh weight, dry leaf weight, and total dry weight of hydroponic lettuce (Figure 2), contrasting slightly with the results of Lara, *et al.* (2023) who found that a high proportion of nitrate in the nutrient solution surpassed treatments with a low proportion of nitrate in dry weight of leaves and roots. However, a PPFD of 400 μmol·m⁻²·s⁻¹ resulted in 55 % greater fresh leaf weight (Figure 2A), 61 % greater dry leaf weight (Figure 2B), and 63 % greater total dry weight (Figure 2C) compared to weights obtained with 200 μmol·m⁻²·s⁻¹.

This weight gain is likely due to increased photosynthetic activity under higher light intensity. These results are consistent with those reported by Avendaño *et al.* (2020) and Johkan *et al.* (2012), who indicated that increasing light intensity from 127 to 235 μmol·m⁻²·s⁻¹ and from 100 to 300 μmol·m⁻²·s⁻¹ respectively increases biomass and weight in hydroponic lettuce. They also agree with Miao *et al.* (2023) and Jin *et al.* (2023), who report that gradually increasing the light intensity to 300 μmol·m⁻² ·s⁻¹ has a positive effect on the fresh and dry weight of lettuce, resulting in a higher dry matter content, making it necessary to optimize LED light in plant species in order to have a successful crop.

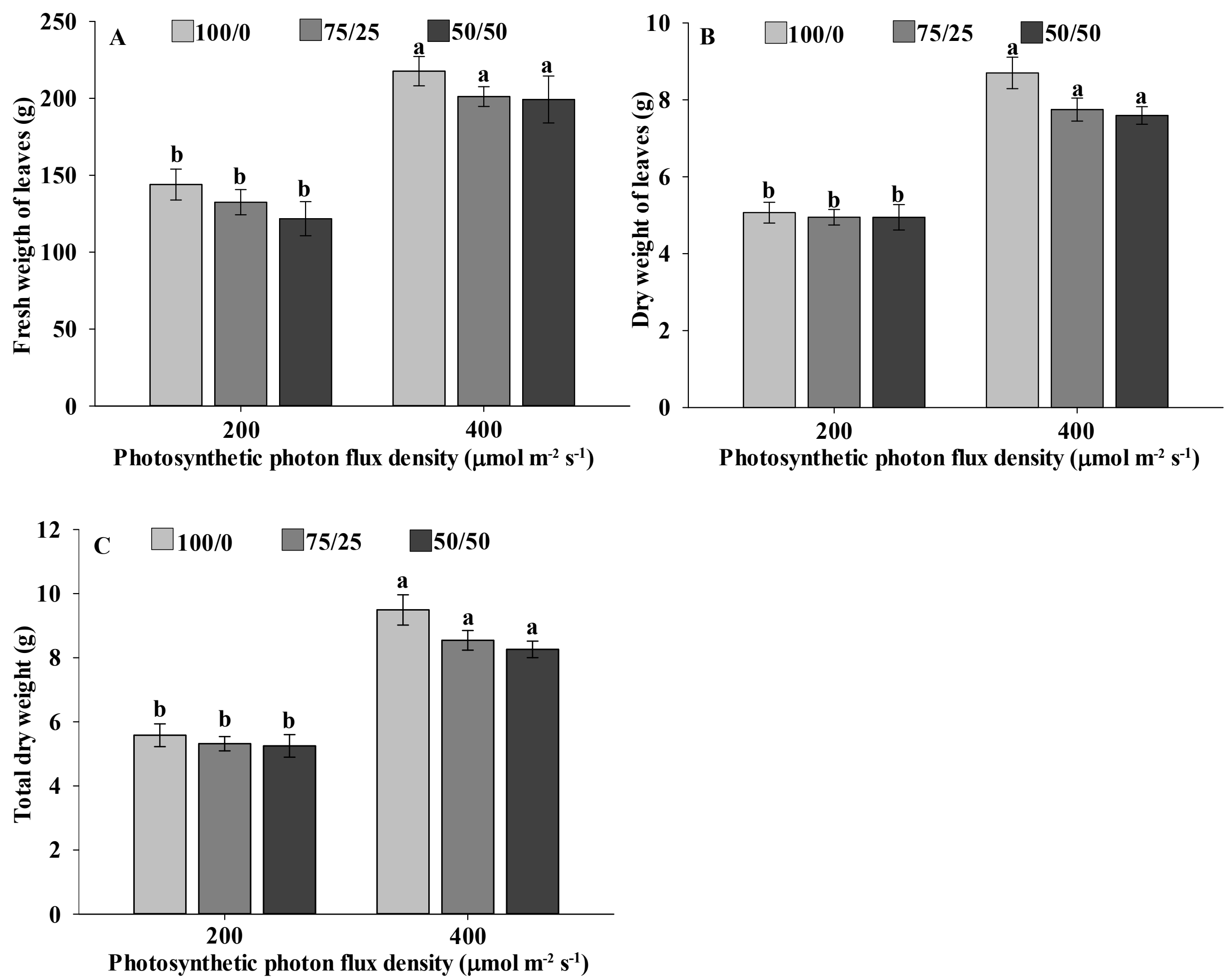

Figure 2. Effect between the nitrate/ammonium ratio in nutrient solution (100/0, 75/25, and 50/50) and photosynthetic photon flux density (200 and 400 μmol·m⁻²·s⁻¹) on fresh leaf weight (A), dry leaf weight (B), and total dry weight per lettuce plant (C). Means ± standard error with identical letters above the bars indicate no significant difference (Tukey, $p\leq0.05$).

Concentration of phosphorus, potassium, calcium, and magnesium in leaves. The nitrate/ammonium ratio did not significantly affect ($p\leq0.05$) the nutritional contents of phosphorus and magnesium in lettuce leaves, but it did have a significant impact on the potassium content. Calcium content was significantly affected at 200 μmol·m^{-2}·s^{-1} PPFD but not at 400 μmol·m^{-2}·s^{-1} PPFD (Table 2). At a 100/0 nitrate/ammonium ratio, potassium and calcium concentrations were 6.18 % and 0.79 %, respectively, which were higher compared to the 50/50 ratio (4.32 % and 0.61 %), representing a decrease of 30 % in potassium and 23% in calcium content.

The reduction in calcium and potassium concentrations in hydroponic lettuce leaves can be attributed to the increased ammonium concentration in the nutrient solution, which leads to a compensatory decrease in calcium and potassium to maintain the overall cation balance (Steiner, 1984). Therefore, the reduction of calcium and potassium in the nutrient solution decreases the concentrations of these elements in lettuce leaves (Mengel and Kirkby, 2000). This result is consistent with Chaillou and Lamaze (2001), who noted that ammonium absorption depolarizes cell membranes, thereby reducing the uptake of potassium and calcium. Similarly, Parra *et al.* (2012) reported that the nitrate/ammonium ratio directly influenced calcium concentration in tomato leaves and stems, although the effect on potassium was less pronounced.

Table 2. Phosphorus, potassium, calcium, and magnesium content in dry lettuce leaves by effect of the nitrate/ammonium ratio, PPFD, and their interaction, in a hydroponic production system and white LED lighting.

Factor	Phosphorus	Potassium	Calcium	Magnesium
	%			
NO_3/NH_4				
100/0	0.27 ± 0.02a	6.18 ± 0.19a	0.79 ± 0.07a	0.32 ± 0.03a
75/25	0.25 ± 0.01a	4.69 ± 0.25b	0.62 ± 0.03b	0.34 ± 0.02a
50/50	0.28 ± 0.01a	4.32 ± 0.21b	0.61 ± 0.03b	0.36 ± 0.01a
[1]PPFD				
200	0.28 ± 0.01a	5.09 ± 0.25a	0.77 ± 0.05a	0.38 ± 0.01a
400	0.26 ± 0.01a	5.03 ± 0.27a	0.59 ± 0.02b	0.30 ± 0.02b
NO_3/NH_4 x PPFD				
100/0 x 200	0.28 ± 0.03a	6.23 ± 0.29a	0.95 ± 0.09a	0.37 ± 0.03a
75/25 x 200	0.25 ± 0.01a	4.23 ± 0.32bc	0.65 ± 0.05b	0.39 ± 0.02a
50/50 x 200	0.32 ± 0.01a	4.83 ± 0.21bc	0.70 ± 0.03b	0.39 ± 0.01a
100/0 x 400	0.27 ± 0.02a	6.13 ± 0.28a	0.63 ± 0.06b	0.28 ± 0.04a
75/25 x 400	0.26 ± 0.02a	5.16 ± 0.30ab	0.60 ± 0.03b	0.29 ± 0.03a
50/50 x 400	0.25 ± 0.01a	3.80 ± 0.23c	0.53 ± 0.03b	0.33 ± 0.02a

[1]PPFD = photosynthetic photon flux density (μmol·m^{-2}·s^{-1}). Means ± standard error with identical letters within each column are not statistically different (Tukey, $p \leq 0.05$).

In contrast, PPFD significantly affected calcium and magnesium concentrations (Table 2); at 400 μmol·m^{-2}·s^{-1}, there was a decrease in calcium and magnesium by 23 % and 21 %, respectively, compared to 200 μmol·m^{-2}·s^{-1}. These results concur with those reported by Song *et al*. (2020), who inferred that high light intensity reduces the accumulation of calcium and magnesium in lettuce. They attributed the reduction in these minerals to increased photosynthetic activity, resulting in lower accumulation in plant organs.

Nitrate and ammonium concentration in lettuce leaves: The nitrate/ammonium ratio significantly affected nitrate content in plant tissue (Table 3). At a 50/50 ratio, nitrate concentration in lettuce leaves was 36 % lower than at the 100/0 ratio. This finding is consistent with Lara *et al.* (2019), who reported that ammonium nutrition can reduce nitrate accumulation in leaves. Regarding ammonium, increasing the cation concentration in the nutrient solution with a 50/50 ratio compared to a 100/0 ratio resulted in an 89 % increase in ammonium concentration in the tissue of lettuce leaves (Table 3).

In contrast, the PPFD of 200 μmol·m^{-2}·s^{-1} induced a nitrate concentration in lettuce leaves that exceeded the concentration found in leaves grown under 400 µmol·m⁻²·s⁻¹ by 22.5 %. This result relates to the findings reported by Song *et al.* (2020), who observed higher foliar nitrate concentrations in lettuce produced under light intensities of 250 to 300 µmol·m⁻²·s⁻¹, which decreased when PPFD was increased to 450 µmol·m⁻²·s⁻¹. Conversely, the ammonium cation exhibited the opposite trend; at a PPFD of 200 µmol·m⁻²·s⁻¹, there was a 40 % reduction in ammonium concentration compared to lettuce leaves exposed to 400 µmol·m⁻²·s⁻¹ of PPFD.

Table 3. Nitrate and ammonium content in lettuce leaves affected by the nitrate/ammonium ratio, PPFD, and their interaction in a hydroponic production system with white LED lighting.

Factor	Nitrate	Ammonium
	----------mg·kg^{-1} ----------	
NO_3/NH_4		
100/0	1249 ± 53.34a	1377 ± 222.77b
75/25	833 ± 59.82b	2416 ± 136.96a
50/50	797 ± 63.82b	2607 ± 180.44a
[1]PPFD		
200	1057 ± 41.88a	1600 ± 166.70b
400	863 ± 81.29b	2667 ± 128.94a
NO_3/NH_4 x PPFD		
100/0 x 200	1182.44b ± 99.07ab	678.76 ± 52.71c
75/25 x 200	1013.36c ± 51.06b	2050.19 ± 56.35b
50/50 x 200	974.60d ± 24.05b	2071.04 ± 151.21b
100/0 x 400	1315.18a ± 30.68a	2075.67 ± 143.00b
75/25 x 400	652.10e ± 12.33c	2782.24 ± 160.54b
50/50 x 400	619.49f ± 68.80c	3143.63 ± 73.11a

[1]PPFD=photosynthetic photon flux density (µmol m^{-2} s^{-1}). Means ± error with identical letters within each column are not statistically different (Tukey, p≤0.05).

.CONCLUSIONS

The incorporation of ammonium into the nutrient solution effectively reduced nitrate concentration while increasing ammonium concentration in lettuce leaves, particularly when combined with a PPFD of 400 µmol·m⁻²·s⁻¹. This finding suggests that ammonium plays a crucial role in modulating nutrient absorption and accumulation in lettuce, which could have significant implications for improving the nutritional quality of hydroponically grown lettuce.

Furthermore, increasing the PPFD to 400 µmol·m⁻²·s⁻¹ significantly enhanced the fresh and dry weight, root dry weight, and total dry weight of lettuce leaves. This demonstrates that higher light intensity promotes greater biomass production, likely due to increased photosynthetic activity. Such results highlight the importance of light management in optimizing growth conditions for hydroponic lettuce.

The addition of ammonium to the nutrient solution, coupled with increased light intensity, presents a viable strategy to boost the biomass of lettuce plants while reducing their nitrate content. This dual approach not only improves the overall yield but also enhances the safety and nutritional value of the produce, addressing both economic and health considerations in lettuce cultivation.

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