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Influence of hydroponic solution based foliar application on yield and quality of mung bean microgreens

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Abstract

This study investigates the impact of various foliar nutrient solutions on the yield and quality of mung bean microgreens to identify the most effective formulation for enhancing growth and nutritional content. The experiment was carried out comparing four modified hydroponic foliar nutrient solutions (NSI): NSI, NSI+MSG (monosodium glutamate), NSI+U (urea) and NSI+AS (ammonium sulphate), alongside a distilled water (DW) control on plant growth and pigment, nitrogen, protein and amino acid composition. Results indicated that NSI treatment significantly improved fresh weight while NSI+MSG showed no significant from NSI and DW. Moreover, NSI+MSG and NSI+AS treatments yielded the highest chlorophyll A and B contents, enhancing nutritional value. Carotenoid contents increased notably with the NSI+AS, NSI+MSG, and NSI+U treatments. The study found significant variations in nitrate, nitrite, and ammonium content, with safe nitrate contents maintained across all treatments. Protein content was highest in the NSI and NSI+MSG treatments, highlighting their potential to enhance microgreen nutritional quality. Essential amino acids such as tyrosine and tryptophan were present across treatments, with phenylalanine detected only in NSI and NSI+MSG. Cysteine was not detected in NSI; only mung bean microgreens treated with NSI+MSG synthesized all four amino acids. In conclusion, NSI+MSG emerges as a promising foliar nutrient solution for optimizing both yield and quality of mung bean microgreens. These findings underscore the importance of tailored nutrient management in microgreen production, offering insights for sustainable agriculture and food security initiatives.

Keywords: Ammonium sulfate, Chlorophyll, Monosodium glutamate, Nitrate, Nitrite, Urea

1. Introduction

Microgreens are immature green vegetables or young seedlings of the herbs. They can be harvested 7 to 21 days after germination and normally when they have developed their first pair of true leaves [1]. They are abundant in flavor, vitamins, minerals, and antioxidants [1]. The microgreens market has grown significantly in the restaurant industry because they serve as attractive garnishes to elevate diverse cuisines, or direct consumption by consumers as fresh additions to salads, soups, sandwiches, and various dishes. Additionally, microgreens are well-suited for urban agriculture, making them an ideal product due to their ability to thrive in a home garden or even a compact space with adequate lighting. A variety of vegetables can be cultivated as microgreens, including broccoli, Chinese kale, purple radish, radish, rat-tailed radish, red cabbage, fenugreek, green pea, lentil, black sesame, buckwheat, morning glory, red roselle, sunflower, and mung bean [2].

Mung beans are staples in whole seed, flour, and especially sprout forms which are used in countless Thai meals [3]. However, in recent years, consumers in the cities have started to incorporate mung bean microgreens into their diets for convenience and health purposes [2]. The increasing consumption of microgreens shows the

change in diet culture, across the globe and within the country, as these products are known to be hyper nutritious and far more healthy in comparison to regular mung bean sprouts [2]. Mung bean microgreens are abundant in carbohydrates, protein, fats, calories, chlorophyll, carotenoids, and phenolic compounds, and display significant antioxidant activity. Mung bean microgreens also exhibit high levels of essential minerals like potassium, phosphorus, calcium, magnesium, sodium, iron, zinc, copper, and magnesium [4]. Furthermore, mung bean microgreens are rich in amino acids such as alanine, arginine, asparagine, aspartic acid, citrulline, cysteine, ɣ-aminobutyric acid, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, and valine [4].

Hydroponic systems can be used to provide tailored levels of nutrients necessary for plant development [5]. Complementary to root uptake, foliar application of nutrients can be directly absorbed through the leaves and assist growth during the initial, more critical phases of microgreen development. An example includes the use of monosodium glutamate (MSG) as a fertilizer to help with growth and plant physiological responses [6]. A foliar application of a nutrient solution with N-P-K ratios of 20-8.7-16.6 at 100 mg/L in the first few days after planting has been reported to enhance the fresh weight of several species of microgreens significantly [7]. These findings indicate that specific foliar nutrition can optimize plant potential yields, although the evidence is inconsistent between different species [4].

In addition, plant leaves can absorb nutrients through foliar nutrition, which plays a crucial role by enabling the easy and rapid absorption of nutrients through the stomata of the leaf cuticle to reach the cells [8]. In recent times, considerable research attention has been directed towards hydroponic microgreens cultivation. In the hydroponic system, the growth and quality of microgreens derived from basil, Swiss chard, and rockets have shown enhanced yields and improved overall quality [9]. When cultivated on vermicompost and in a hydroponic system, cabbage microgreens exhibit enhanced nutritional values in comparison to their mature counterparts. These improvements encompass important trace minerals essential for dietary intake, including phosphorus (P), potassium (K), sulfur (S), calcium (Ca), magnesium (Mg), manganese (Mn), copper (Cu), zinc (Zn), iron (Fe), and sodium (Na) [10].

Nitrogen is vital for many cell compounds like chlorophyll, amino acids, enzymes (e.g., ribulose-1,5-bisphosphate carboxylase-oxygenase), and hormones, and plays a crucial role in plant growth and development [11]. Plants have the ability to assimilate nitrogen as an essential macronutrient in several different forms, including inorganic forms (nitrate and ammonium) and organic forms (such as urea, glutamate, and various free amino acids) to support plant growth [12]. Nitrate is among the nitrogen sources that plant roots can assimilate and then transfer to the shoots, where it can be either reduced or stored within vacuoles. This form of nitrogen plays a pivotal role in affecting plant photosynthetic capacity, growth, and crop yield [13]. Ammonium is an alternate form of nitrogen employed in agriculture to support the growth and development of plants. Typically, plants must convert nitrate into ammonium before combining it into organic compounds [14]. In terms of nitrogen fertilizers, urea, the most widely utilized nitrogen source in agriculture, can be directly absorbed by plants or metabolized into ammonium or nitrate by soil microorganisms after degradation [15]. Recently, monosodium glutamate has emerged as an alternative nitrogen fertilizer, playing a role in stimulating plant growth [16].

Most microgreens are cultivated on growing media with minimal or no initial fertilizer. Therefore, foliar nutrition becomes a suitable, cost-effective, and labor-efficient choice. Foliar-nutrient solutions have been documented to enhance fresh biomass while their effect on dry weight is limited, as seedlings need fewer nutrients in their early stages of growth [6]. However, foliar applications have much greater potential to affect physiological attributes, especially pigments. There has been documentation for some microgreen species that the additional nutrients promote the increase of chlorophyll A [17] and that biostimulant foliar sprays have more effect than distilled water on radish microgreens [18]. Other crops have more documented fortified pigment enhancement, such as ammonium sulfate that is more effective on chlorophyll B of spring wheat than ammonium nitrate or urea [19]. Likewise, in sunflower microgreens, foliar-applied urea resulted in the highest xanthophyll content compared to other nitrogen sources such as ammonium phosphate, potassium nitrate, calcium nitrate, and ammonium sulfate [20]. Moreover, among different nitrogen sources, foliar-applied urea on sunflower microgreens attained the highest xanthophyll content while ammonium phosphate, potassium nitrate, calcium nitrate, and ammonium sulfate were the other sources [21]. Therefore, enhancing the productivity and quality of microgreens can be achieved through the foliar application of a customized Hoagland's nutrient solution. Accordingly, this study examined the foliar application of various nutrient solutions derived from a modified Hoagland's nutrient solution, aiming to enhance both the yield and quality of mung bean microgreens.

2. Materials and methods

2.1 Plant growth conditions

Mung bean seeds (*Vigna radiata* L.) were individually weighed at 50 g and soaked in warm water at 50°C for 6 hours. Tissue papers were used as a substrate to support root growth and layered onto a plastic tray (33x25x5

cm) with drain holes. The trays were moistened with distilled water until the papers were thoroughly wet before sowing. After sowing, the trays were kept at room temperature (28-30°C) and irrigated with 100 mL of distilled water per tray daily for 5 days. Following this period, the microgreens were treated with a foliar application of different fertilizers (30 mL/tray) for each treatment and allowed to stand for 24 hours [22].

Subsequently, these treated microgreens were placed under a red-blue LED light with a photosynthetic photon flux density (PPFD) of 250 µmol/m^2/s for 48 h before being harvested. Each treated microgreens were washed with distilled water, then placed into resealable plastic bags and stored in a freezer at -4°C, awaiting further analysis. The study was carried out at the Laboratory of Urban Agriculture Technology, Division of Agricultural Technology, Department of Agricultural and Fishery Science, Faculty of Science and Technology, Prince of Songkla University, Pattani Campus.

2.2 Foliar nutrient preparation for the treatments

The study included five foliar treatments and the foliar nutrient solutions were based on a modified Hoagland's hydroponic nutrient solution as described by Sirinupong (2017) [23]. This hydroponic solution had named Nutrient Solution I (NSI), consisted of potassium nitrate (390 mg/L), monoammonium phosphate (65 mg/L), calcium nitrate (500 mg/L), magnesium sulfate (250 mg/L), monopotassium phosphate (50 mg/L), manganese sulfate (4 mg/L), microelements (Boron EDTA, MnEDTA, MgO, CuEDTA, MoEDTA, and FeEDTA) (4 mg/L), and iron chelate (25 mg/L). Five foliar treatments were prepared as followed: (1) NSI (Nutrient Solution I); (2) NSI+AS (NSI with 1.316 mg/L of Ammonium Sulfate); (3) NSI+U (NSI with 2.561 mg/Lof Urea); (4) NSI+MSG (NSI with 3.365 mg/L of Monosodium Glutamate); and (5) DW (Distilled Water), used as control treatment.

2.3 Fresh and dry weight determination

To measure fresh weight, mung bean microgreens were gently blotted with a piece of soft tissue paper to eliminate any excess surface moisture. Subsequently, 100 microgreen seedlings were promptly weighed, and the data was collected. To determine dry weight, 100 mung bean microgreen seedlings were dried overnight in an oven at 65°C for 48 h, and then the samples were weighed.

2.4 Chlorophylls, carotenoid and xanthophyll determination

The analysis of chlorophyll A, chlorophyll B, total chlorophyll, carotenoid, and xanthophyll contents followed the extraction procedures outlined by Duma et al (2014) [24]. Briefly, 0.1 g of fresh microgreens were precisely weighed and ground using a mortar and pestle, then extracted with 15 mL of ethanol. Subsequently, the extracted solution was filtered through filter paper No. 1 and transferred into a microcentrifuge tube. These tubes were kept in darkness and then subjected to centrifugation before being transferred into a 96-well microplate. Chlorophyll, carotenoid, and xanthophyll contents were determined spectrophotometrically through absorption measurements ranging from 350 to 700 nm, and were calculated using the following equations:

$$\text{Chloroplyll a (mg/g FW)} = \frac{13.7A_{665} - 5.76A_{649}}{mass \times 200} \quad (1)$$

$$\text{Chloroplyll b (mg/g FW)} = \frac{25.8A_{649} - 7.6A_{665}}{mass \times 200} \quad (2)$$

$$\text{Carotenoids (mg/g FW)} = \frac{4.7A_{440} - 0.263c_{chla+chlb}}{mass \times 200} \quad (3)$$

$$\text{Xanthophyll (mg/g FW)} = \frac{11.51A_{480} - 20.61A_{495}}{mass \times 200} \quad (4)$$

2.5 Nitrate, nitrite and ammonium determination

The nitrate content was measured through a colorimetric method using salicylic acid [25]. Firstly, 2 g of fresh microgreen samples were extracted with 5 mL of deionized water, followed by incubation in a boiling water bath for 20 min. Once the mixture had cooled to room temperature, the supernatant was obtained by centrifugation at 20,100 x g at 4°C for 10 min. A 10 µL aliquot of the extract was mixed with 40 µL of a 0.05% (w/v) salicylic acid solution in sulfuric acid in a 1.5 mL microtube, and vortexed vigorously. The mixture was then allowed to stand at room temperature for 20 min, followed by the addition of 1 mL of 8% (w/v) NaOH solution in deionized water.

The spectrophotometer was utilized to measure the absorbance at 410 nm. The nitrate content (µmol/g of fresh weight) for each sample was then calculated by the following formula:

$$[\text{True nitrate concentration (mM)}] = [\textit{extracted volume (ml)}] / [\textit{fresh weight (g)}] \quad (5)$$

The nitrite content was evaluated using the Griess reaction method [25]. Firstly, 2 g of microgreen samples were extracted using 5 mL of deionized water. The supernatant was obtained by centrifugation at 20,100 x g at 4°C for 10 min. Subsequently, 260 µL of the supernatant was mixed with 65 µL of 1% (w/v) sulfanilamide in 1 mol/L of hydrochloric acid, 65 µL of 0.02% (w/v) N-1-naphthylethylenediamine dihydrochloride in deionized water, and 910 µL of deionized water in 1.5 mL microtube. The mixture was then kept at room temperature for 15 min for incubation. Subsequently, the absorbance was measured at 540 nm using a spectrophotometer. The nitrite content for each sample in µmol/g of fresh weight was determined using the following formula:

$$[\text{True nitrite concentration (µM)}] = [\textit{extracted volume (mL)}] / [\textit{fresh weight (g)}] \quad (6)$$

The ammonium content was determined using the ammonia-salicylate method [25]. Initially, 2 g of the microgreen sample was extracted with 5 mL of 0.1 M potassium chloride, and the supernatant was obtained by centrifugation at 20,100 x g at 4°C for 10 minutes. A 40 µL aliquot of the supernatant was carefully transferred into a 96-well plate, followed by the addition of 80 µL of the salicylate/nitroprusside solution and 80 µL of the hypochlorite solution. The mixtures were allowed to incubate at room temperature for 45 min, after which the absorbance was measured at 650 nm using a spectrophotometer. The apparent ammonium concentration (mM) of the supernatant was determined using a standard curve, and the ammonium content of the sample (in µmol/g fresh weight) was calculated using the following formula:

$$[\text{True ammonium concentration (µM)}] = [\textit{extracted volume (mL)}] / [\textit{fresh weight (g)}] \quad (7)$$

The salicylate/nitroprusside solution was prepared by dissolving 150 g of sodium salicylate and 0.30 g of sodium nitroprusside and making up the volume to 1 L, while the hypochlorite solution was made by diluting 6 mL of 5.25% sodium hypochlorite to a final volume of 100 mL.

2.6 Protein and amino acid content determination

To determine the protein content and estimate amino acid compositions, the microgreens were first dried and then ground into a powder using a mortar and pestle. A ground sample weighing 1.25 g was measured and transferred into a 50 mL beaker. The sample was extracted using 10 mL of methanol, which was added and warmed on a hotplate with stirring for 5 min. The mixture was then allowed to stand for 15 min. The solution was filtered through filter paper No. 1 into a 25 mL volumetric flask, and the volume was adjusted with methanol. The absorbance was measured within the wavelength range of 200 to 600 nm using a UV-Vis spectrophotometer [26]. Different amino acids were identified by their specific wavelengths: cysteine (204–220 nm), phenylalanine (240–265 nm), tyrosine (274–330 nm), tryptophan (275–312 nm), and histidine (above 312 nm) [26]. The protein concentration was calculated using a percent solution extinction coefficient, typically ranging from 4.0 to 24.0, with an average of approximately 10 for a mixture of various proteins. The protein concentration was calculated using the following formula:

$$\text{Concentration in mg/mL} = (\textit{Absorbance} / {}^{\varepsilon}\textit{percent}) \times 50 \quad (9)$$

To calculate the results on a weight basis, the concentration obtained (mg/mL) was multiplied by the final extract volume (25 mL) and the sample mass (1.25 g) was taken into consideration. Therefore, the values for protein and amino acids were documented as:

$$\text{Protein content (mg/g dry weight)} = \textit{Concentration (mg/mL)} \times 25 / 1.25 \quad (9)$$

2.7 Statistical analysis

In this experiment, we used five treatments and three replications with a completely randomized design (CRD). Data processing was conducted with MS Excel software, version 7.0. Means marked with different letters indicated significant differences at a 95% confidence level, as determined by the Least Significant Difference (LSD) test.

3. Results and discussion

3.1 Plant growth and yield

In this research, we investigated the most suitable foliar nutrient solution among various formulations: NSI, NSI+MSG, NSI+U, and NSI+AS. Additionally, distilled water (DW) was used as a control treatment to assess the impact of various foliar applications on mung bean microgreens. The fresh weights of mung bean microgreens receiving the NSI treatment (0.157 g/seedling) showed a slightly higher fresh weight compared to those treated with NSI+MSG (0.155 g/seedling). However, statistical analysis revealed that this difference was not significant (Figure 1A). Conversely, the NSI+U (0.145 g/seedling) and NSI+AS (0.141 g/seedling) treatments significantly reduced the fresh weight of the microgreens compared to the DW control treatment (0.150 g/seedling) exhibited distinctions depending upon the application of different foliar nutrient solutions. Specifically, NSI and NSI+MSG had the greater potential to improve fresh weights. Hydroponic nutrition can provide optimal nutrient levels for plant growth [17], and MSG can be used as a fertilizer to improve the plant growth [18]. Additionally, it has been reported that applying a foliar nutrient solution with a composition of 20-8.7-16.6 (N-P-K) at a concentration of 100 mg/L for four days after planting increased the fresh weight of ten microgreen species [19]. Suitable foliar nutrient solutions have the capacity to enhance microgreen yields, and different plant species may require specific conditions to maximize their yield potential [2].

For dry weight, the NSI+MSG treatment (0.0153 g/seedling) resulted in the highest dry weight among the treatments, followed by NSI+U, NSI+AS, NSI, and DW treatments, with dry weights of 0.0145, 0.0143, 0.0138, and 0.0118 g/seedling, respectively (Figure 1B). However, the differences in dry weight among these treatments were not statistically significant. This finding is consistent with previous observations in sunflower microgreens [13]. Similarly, it has been reported that the influence of foliar nutrient solutions had a relatively minor impact on dry weight, primarily because plants utilize fewer nutrients during the seedling stage [27].

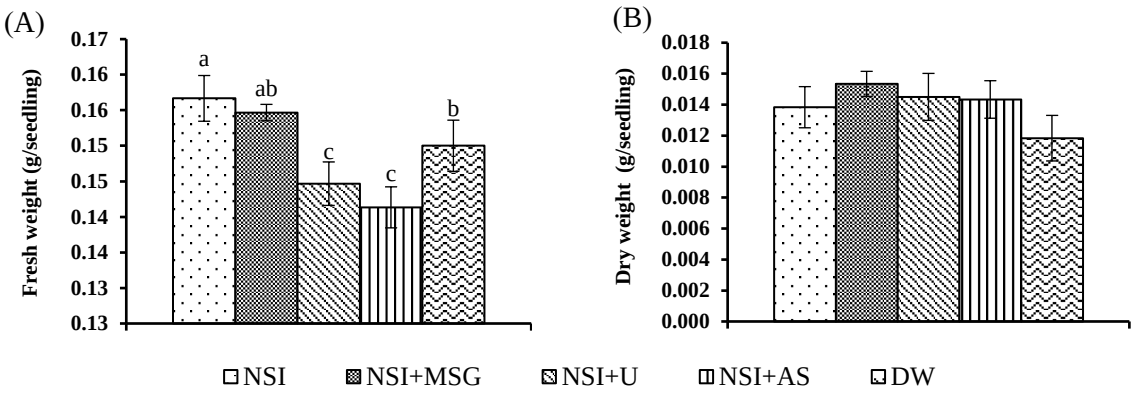

Figure 1 Fresh weight (A), and dry weight (B) of mung bean microgreens under various foliar nutrient solution applications. Distinct letters within each column plot indicate significant variations at the 95% confidence level, based on the LSD (n=15), and the bars represent the standard deviations.

3.2 Chlorophyll A, chlorophyll B, total chlorophyll, carotenoid and xanthophyll content

There was statistically significant variation in chlorophyll A, chlorophyll B, total chlorophyll, xanthophyll and carotenoid content among different foliar treatments in mung bean microgreen (Figure 2). Chlorophyll, the green pigment responsible for photosynthesis in plants, plays a crucial role in the growth, indicators of freshness, and nutritional value and quality of microgreens [21]. Mung bean microgreens treated with NSI+AS and NSI+MSG (both 0.42 mg/g FW) had the highest chlorophyll A content, significantly higher than those treated with NSI+U, NSI, and DW (Figure 2A). It has been revealed that different plant species of microgreens show different chlorophyll A contents, and its content was increased after supplying a nutrient solution [22]. Moreover, it has been reported biostimulant solutions increased chlorophyll A in radish microgreens more effectively than distilled water [23]. Similarly, NSI+AS treatment produced the highest Chlorophyll B content (0.29 mg/g FW) followed by NSI+U, NSI+MSG, NSI, and DW treatments with Chlorophyll B contents of 0.26, 0.25, 0.20, and 0.09 mg/g FW, respectively (Figure 2B). Similar findings were observed in spring wheat plants, where ammonium sulfate increased chlorophyll B content more than ammonium nitrate and urea [24]. Furthermore, the study found that mung bean microgreens treated with foliar nutrient solutions such as NSI+AS (0.71 mg/g FW), NSI+MSG (0.66 mg/g FW), and NSI+U (0.66 mg/g FW) exhibited significantly higher total chlorophyll contents compared to the DW treatment (0.45 mg/g FW) (Figure 2C). These findings suggest that various foliar application of nitrogen

compounds can have a significant positive impact on the total chlorophyll, chlorophyll A and B content, in which NSI+AS and NSI+MSG providing the highest levels among the foliar nutrient solution.

However, xanthophyll content was highest in mung bean microgreens treated with NSI and it was not statistically different from NSI+MSG and DW treatments. In contrast, these treatments exhibited significantly higher xanthophyll content, approximately double compared to the NSI+U treatment. It is important to note that data for xanthophyll content was unable to detect for the NSI+AS treatment (Figure 2D). These results suggest that nutrient composition significantly impacts xanthophyll accumulation. Interestingly, foliar application of urea fertilizer to sunflower microgreens resulted in the highest xanthophyll content compared to ammonium phosphate, potassium nitrate, calcium nitrate, and ammonium sulfate [6]. This discrepancy highlights that the influence of foliar nutrient solutions on xanthophyll content can vary depending on the specific nutrient composition and the microgreen variety used. It is crucial to consider these factors when optimizing nutrient solutions for different microgreens to enhance their nutritional quality.

Carotenoid content showed the highest after using foliar application of the NSI+AS treatment (0.26 mg/g FW). However, this treatment was not statistically significant when compared to the carotenoid content in the NSI+MSG treatment (0.25 mg/g FW). Nonetheless, both of these treatments exhibited significantly higher carotenoid content in comparison to the NSI+U, DW, and NSI treatments, which recorded 0.24, 0.16, and 0.15 mg/g FW, respectively (Figure 2E). Carotenoids are important for their antioxidant properties and contribution to plant health and nutritional quality [25]. The significant increase in carotenoid content with the NSI+AS, NSI+MSG, and NSI+U treatments suggests that these nutrient solutions provide a more balanced and effective nutrient supply such as nitrogen and magnesium that promotes carotenoid biosynthesis [26].

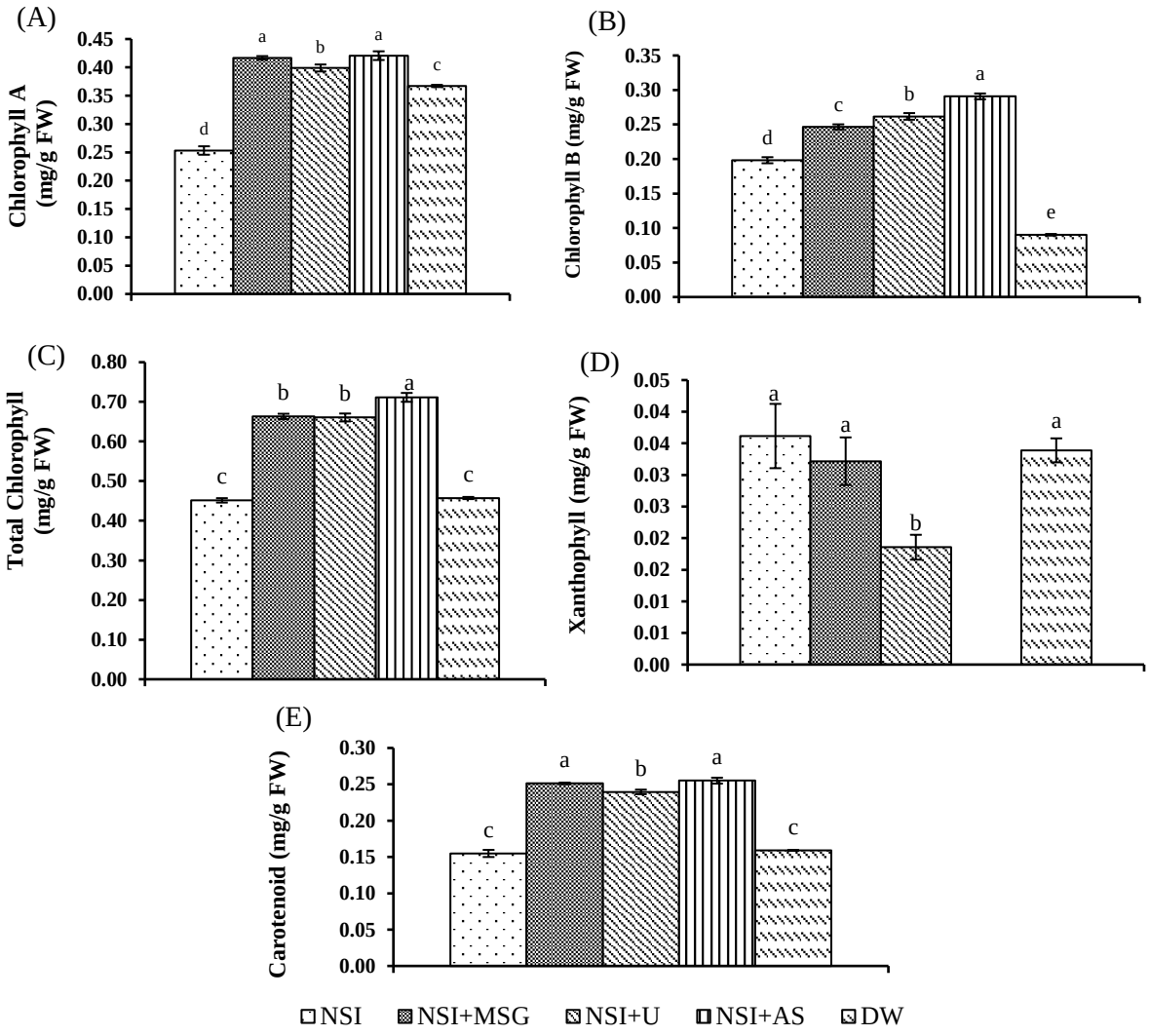

Figure 2 Chlorophyll A content (A), chlorophyll B content (B), total chlorophyll content (C), xanthophyll content (D), and carotenoid content (E) of mung bean microgreens under various foliar nutrient solution applications. Distinct letters within each column plot indicate significant variations at the 95% confidence level, based on the LSD (n=15), and the bars represent the standard deviations.

3.3 Nitrate, nitrite, ammonium, and protein contents

Nitrate serves as a vital nitrogen source for plants, essential for synthesizing proteins, enzymes, and other crucial molecules. According to the World Health Organization and the European Food Safety Authority, the acceptable daily intake of nitrate and nitrite is 3.7 mg/kg body weight and 0.07 mg/kg body weight, respectively [28]. However, excessive nitrate accumulation can pose health risks due to metabolites like nitrite, nitric oxide, and N-nitroso compounds, which are associated with human disorders such as methemoglobinemia [27]. Among these treatments, NSI+MSG displayed the highest contents at 0.172 μmol/g FW, followed by NSI+AS, NSI, NSI+U, and DW, which exhibited contents of 0.172, 0.160, 0.149, 0.149, and 0.137 μmol/g FW, respectively (Figure 3A). However, there is no statistically significant differences in nitrate content among the various foliar application treatments and distilled water (Figure 3A), suggesting their suitability as valuable dietary sources for both humans and plant growth. This finding aligns with observations in cauliflower microgreens but contrasts with micro broccoli raab and micro broccoli microgreens, where different ammonium ratios led to significant variations [20].

Conversely, nitrite content displayed an opposite trend compared to nitrate content. The highest nitrite content was observed in the DW treatment (26.98 μmol/g FW), which did not significantly differ from the NSI treatment (21.98 μmol/g FW). Both DW and NSI treatments exhibited significantly higher nitrite levels compared to NSI+MSG (20.10 μmol/g FW), NSI+AS (9.69 μmol/g FW), and NSI+U (8.75 μmol/g FW) treatments (Figure 3B). Nitrite is a naturally occurring compound found in various foods, particularly plant-based foods, and vegetables. It is also utilized as an additive in industrially processed foods. Particularly, processed meat and animal food products are significant sources of dietary nitrite [28].

In natural and agricultural ecosystems, ammonium constitutes a primary source of inorganic nitrogen for plants, even though its concentration in the soil solution can be over three times lower than that of nitrates [29]. In this study, there were significant variations in the ammonium content of the different treatments given to mung bean microgreens. The NSI+U treatment recorded the highest ammonium content (204.27 μmol/g FW), and there was no significant difference when compared to the NSI+AS (183.61 μmol/g FW) treatment. Both treatments showed significant differences compared to the NSI+MSG, NSI, and DW treatments 126.61, 92.27, and 54.94 μmol/g FW, respectively) (Figure 3C).

Moreover, the protein content in mung bean microgreens treated with various foliar applications exhibited significant differences among the treatments. The NSI treatment displayed the highest protein content at 135.97 mg/g, with no statistically significant difference observed when compared to the NSI+MSG treatment (135.87 mg/g). The NSI+U and NSI+AS treatments closely followed, displaying protein contents of 129.07 and 128.87 mg/g, respectively. Importantly, all nutrient treatments showed significantly higher protein content than the DW treatment, which had a protein content of 121.97 mg/g (Figure 3D). Thus, this study highlights the role of targeted nutrient solutions in optimizing microgreen nutritional quality. This enhancement in protein levels, along with the superior nutritional profile of microgreens over mature vegetables, underscores their potential as a valuable dietary source. It has been reported that microgreens have emerged as a notable source of plant protein, boasting higher protein content than both baby greens and mature vegetables [30].

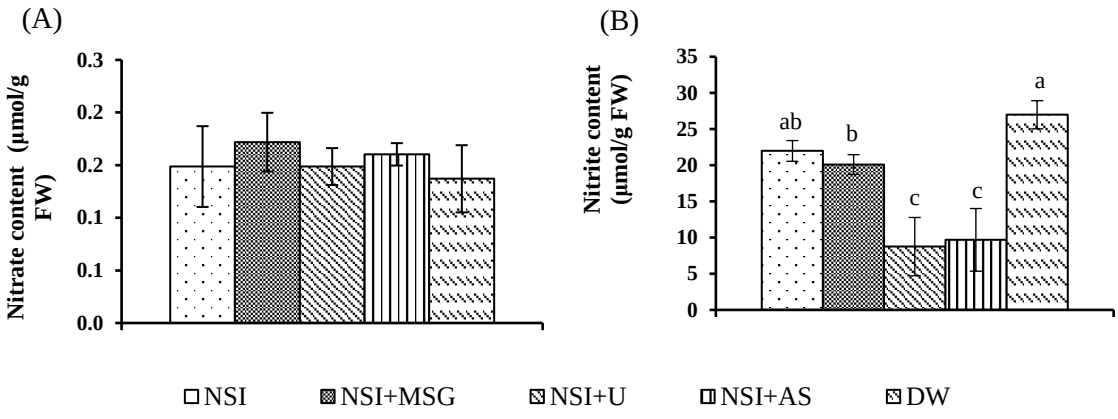

Figure 3 Nitrate content (A), nitrite content (B), ammonium content (C) and protein content (D) of mung bean microgreens under various foliar nutrient solution applications. Distinct letters within each column plot indicate significant variations at the 95% confidence level, based on the LSD (n=15), and the bars represent the standard deviations.

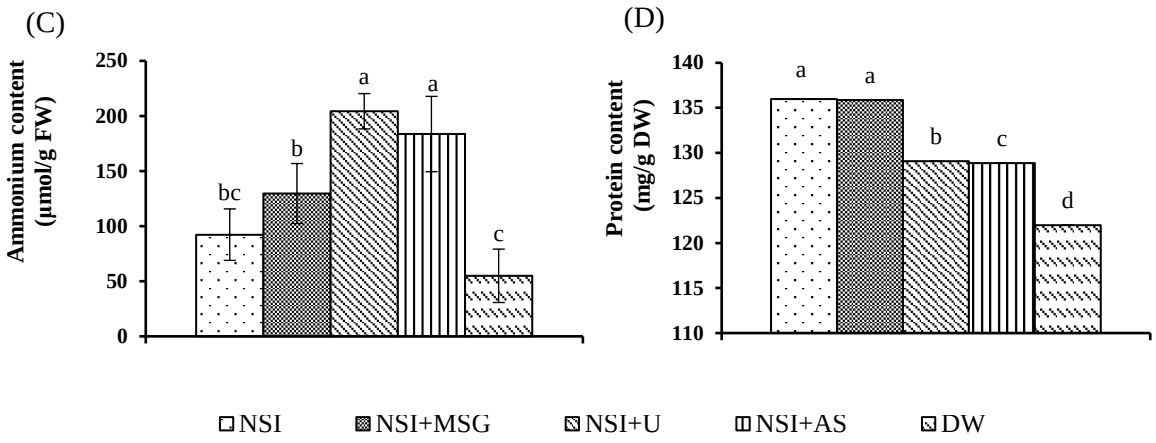

Figure 3 (cont.) Nitrate content (A), nitrite content (B), ammonium content (C) and protein content (D) of mung bean microgreens under various foliar nutrient solution applications. Distinct letters within each column plot indicate significant variations at the 95% confidence level, based on the LSD (n=15), and the bars represent the standard deviations.

3.4 Estimation of amino acid compositions

In this experiment, we conducted an estimation of four specific amino acids (cysteine, phenylalanine, tyrosine, and tryptophan) in mung bean microgreens subjected to foliar application of various nutrition solutions (Table 1). Our findings revealed that cysteine content was absent only in the NSI treatment but it was detectable in microgreens treated with NSI+MSG, NSI+AS, NSI+U, and DW. Cysteine is one of the key ingredients needed for protein synthesis and is categorized as a non-essential amino acid [29]. Wojdylo et al. (2020) verified the presence of cysteine in kale (*Brassica oleracea*) microgreens, while it was not found in mung beans (*Vigna radiata*) microgreens [30].

Phenylalanine is recommended for various medical conditions, including the alleviation of alcohol withdrawal symptoms, treatment of vitiligo, assistance in weight loss, management of depression, relief from rheumatoid arthritis and osteoarthritis, pain management, support for multiple sclerosis, mitigation of symptoms in Parkinson's disease, and addressing attention deficit-hyperactivity disorder [31]. In this study, phenylalanine was found solely in mung bean microgreens that underwent NSI and NSI+MSG treatments, with no presence detected in the other treatments. The previous study by Kaur et al. (2022) also reported a comparable finding where mung bean and black gram microgreens exhibited high levels of phenylalanine [4]. Furthermore, Wojdylo et al. (2020) identified the presence of phenylalanine in various types of sprouts, with mung bean sprouts showing the highest content [32]. Additionally, they also detected phenylalanine in microgreens of kale, radish, green peas, beetroot, and amaranths.

Tyrosine, an amino acid, serves as the precursor to the catecholamine neurotransmitters dopamine and norepinephrine [33]. This studied revealed the presence of tyrosine in mung bean microgreens across all treatments. In the study conducted by Wojdylo et al. (2020), it was observed that various types of sprouts exhibited elevated levels of tyrosine. Conversely, kale, radish, beetroot, green pea, and amaranth microgreens were found to have comparatively lower tyrosine content [30].

Tryptophan has the potential to influence a wide array of diseases and conditions in animal and human health [34]. This study revealed that the tryptophan has been detected in all kind of foliar nutrient application and it is aligned well with the previous study by Wojdylo et al. (2020). They found tryptophan in several types of sprouts (radish, lentil, black medick, broccoli, sunflower, leek, beetroot, and mung bean) and microgreens (kale, radish, beetroot, green pea, and amaranth) [30].

Table 1 Estimation of amino acid compositions (cysteine, phenylalanine, tyrosine, and tryptophan) in mung been microgreens treated with foliar application of different nutrition solutions.

Treatment	Cysteine	Phenylalanine	Tyrosine	Tryptophan
NSI	-	+	+	+
NSI+MSG	+	+	+	+
NSI+U	+	-	+	+
NSI+AS	+	-	+	+
DW	+	-	+	+

4. Conclusions

The application of different foliar nutrient solutions positively impacted the yield and quality of mung bean microgreens. NSI significantly enhanced fresh weight yield, while NSI+MSG improved yield and various quality indicators such as chlorophyll, carotenoid content, and protein levels. Although nitrate and nitrite levels varied, all these foliar nutrient solutions remained safe. Among various foliar nutrition treatments, all essential amino acids investigated in this study were detected only in NSI+MSG treatments. These findings highlight the importance of nutrient management in optimizing microgreen growth and nutritional content, valuable for sustainable agriculture and food security.

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7. Author contributions

ER: Conceptualization, study design, experimentation, data curation, and drafting of the manuscript; PH: assistance with experimentation and data curation; NMPSH: conceptualization, validation, review and editing of the manuscript, and approval of the final version.

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