Hue University Journal of Science: Natural Science
Vol. 134, No. 1S-2, 5–16, 2025 (Special Issue: CBA 2025)

pISSN 1859-1388
eISSN 2615-9678

Effects of Nitrogen Concentration on Growth and Quality of Hydroponically Grown Purple Kale (*Brassica oleracea* var. *sabellica*)

Le Thi Thuy*, Dang Thi Van Anh

Hanoi National University of Education, Hanoi, Vietnam

* Correspondence to Le Thi Thuy <hienthuy20@gmail.com>

(*Received: 27 May 2025; Revised: 23 August 2025; Accepted: 25 August 2025*)

Abstract. This study evaluated the effects of nitrogen concentration in hydroponic nutrient solution on the growth and quality of purple kale (*Brassica oleracea* var. *sabellica*). The standard Knop solution (170.8 mg N/L) was used as the control, and six experimental treatments were established by increasing N levels to 1.5, 2, 2.5, 3, 3.5, and 4 times that of the control. The results showed that, 512.4 mg N/L (T4) promoted the best growth, with a plant height of 36.70 cm, 13.60 leaves, 68.93 g fresh weight, and 2.71 mg/dm^2 chlorophyll at 70 days after sowing. In contrast, 341.8 mg N/L (T2) yielded the best quality, with total organic acid content of 55.61 mg/g, vitamin C content of 58.66 mg/100 g, reducing sugar content of 4.46 mg/g, and anthocyanin content of 0.65 mg/g. Importantly, nitrate content at T2 (785.54 mg/kg fresh weight) remained below the European Union safety threshold. These results provide a scientific basis for optimizing nutrient solutions to balance growth and quality in hydroponic purple kale cultivation.

Keywords: anthocyanin, Knop solution, hydroponic system, nitrogen concentration, purple kale (*Brassica oleracea* var. *sabellica*)

1 Introduction

Purple kale (*Brassica oleracea* var. *sabellica*) is a leafy green vegetable belonging to the family Brassicaceae. It originates from the Mediterranean and Asia Minor regions and is now widely cultivated in temperate and subtropical climates around the world [1]. Kale has been dubbed a "superfood" due to its superior nutritional profile compared to many other vegetables. It is commonly used in health-conscious diets thanks to its richness in vitamins A, B, C, and K, as well as essential minerals such as calcium (Ca), potassium (K), iron (Fe), zinc (Zn), and a wide array of antioxidants including carotenoids and flavonoids [2]. Notably, purple kale contains high levels of anthocyanins, which have the capacity to neutralize free radicals, reduce oxidative stress, protect cells from damage, and lower the risk of cardiovascular diseases and cancer [3, 4, 5]. Given these nutritional and medicinal benefits, investigating and determining optimal nutrient conditions for purple kale cultivation plays a critical role in enhancing both yield and quality of this valuable crop.

Nitrogen is an essential nutrient for plants, playing a critical role in nearly all metabolic and energy conversion processes at the cellular and organismal levels. It forms the structural basis of key organic compounds such as proteins, nucleic acids, chlorophyll, and hormones [6]. Therefore, nitrogen deficiency leads to stunted plant growth, reduced crop quality, and lower yields, especially in leafy vegetables [7, 8]. Plant nitrogen requirements vary depending on species, growth stage, and environmental conditions. Among the various sources, ammonium, nitrate, and urea are the primary forms of nitrogen absorbed by plants

DOI: 10.26459/hueunijns.v134i1S-2.7893

[9, 10]. Research on sweet potato indicated clear nitrogen deficiency symptoms at 0–50 mg/L, with the most vigorous growth observed at 250 mg/L, while the highest tuber yield was recorded at 200 mg/L [11]. Similarly, optimal growth and yield of *Persicaria minor* was achieved at 100 mg/L nitrogen, whereas the highest levels of quercitrin and quercetrin-3-glucuronide were found at 50 mg/L [12]. In hydroponically grown lettuce, increasing nitrogen concentrations significantly improved growth and productivity [13]. Additionally, the form and ratio of nitrogen sources also influence crop performance; for instance, a combination of 75% urea nitrogen and 25% nitrate nitrogen was shown to enhance tomato growth by promoting photosynthesis, sugar accumulation, and enzymatic activity [10]. In practical agriculture, fertilizers—particularly inorganic nitrogen fertilizers—serve as the main nitrogen sources for crops, and their nitrogen content can affect plant metabolism and the synthesis of secondary metabolites [14, 15].

Hydroponics is a cultivation technique that utilizes nutrient solutions instead of soil to supply water and essential nutrients to plants [16]. These nutrient solutions, composed primarily of dissolved macro- and micronutrients, enhance nutrient availability, minimize soil-borne diseases, and thereby contribute to increased yields and shorter crop cycles [13]. Moreover, the ability to precisely adjust the composition, concentration, and ratio of mineral elements in the solution makes hydroponics an effective system for evaluating the specific roles of nutrients [12]. This technique has been widely applied in the production of vegetables, especially leafy greens [10, 17]. Although the importance of nitrogen for plant growth, particularly in leafy vegetables, has been extensively studied, there is still limited research in Vietnam focusing on how varying nitrogen concentrations affect the growth and development of kale under hydroponic conditions. Therefore, this study was conducted to determine the impact of different nitrogen concentrations on the growth and quality of purple kale, with the aim of proposing an appropriate nutrient regime for cultivating this nutritionally valuable crop.

2 Materials and Method

2.1 Materials

F1 hybrid seeds of purple kale, cultivar Rado 214 of Italian origin, were supplied by Rang Dong Seed Company.

The study used Knop nutrient solution as the control formula, prepared according to the description in [18], with the following concentrations of essential nutrients: nitrogen (N) at 170.8 mg/L, potassium (K) at 137.36 mg/L, phosphorus (P) at 56.9 mg/L, magnesium (Mg) at 24.65 mg/L, sulfur (S) at 32.53 mg/L, chlorine (Cl) at 67.63 mg/L, and iron (Fe) at 4.31 mg/L. Six experimental treatments (designated as T1 to T6) were formulated by increasing the nitrogen (N) concentration to 1.5, 2, 2.5, 3, 3.5, and 4 times that of the control, resulting in nitrogen concentrations of 256.2, 341.8, 427.1, 512.4, 597.8, and 683.2 mg/L, respectively. The adjustments were made using NH_4NO_3 to ensure that all other nutrient concentrations remained consistent with those in the control solution.

2.2 Methods

Purple kale seeds were sown in cocopeat pellets, and after 5–6 days of germination, the seedlings were transferred to a static hydroponic system. The experiment included one control and six nitrogen treatment levels, with each treatment arranged in three Styrofoam containers (25 liters per container), each containing five net pots, and two plants per pot. The containers were placed in a shaded net house and arranged in a completely

pISSN 1859-1388
eISSN 2615-9678

randomized design to ensure uniform light conditions. Each treatment was replicated three times. The nutrient solution was monitored and replenished 1–2 times per week to maintain a pH range of 6.0–6.5, using pH adjustment solutions of H_3PO_4 and KOH as needed. Additionally, the nutrient solution was partially replenished once a week when the volume decreased, ensuring that approximately two-thirds of the root system remained submerged throughout the experiment.

To evaluate the effects of nitrogen concentration in hydroponic solutions on the growth performance of purple kale, several growth parameters were analyzed, including plant height (cm), number of leaves per plant (defined as the total number of leaves present at the time of sampling), and fresh weight (g), using standardized measurement protocols [19]. Total chlorophyll content was estimated using a portable chlorophyll meter (SPAD-502 Plus, Konica Minolta), and the values were converted to mg/dm^2 using an appropriate equation. These indicators were assessed at three time points: 30, 50, and 70 days after sowing.

Quality-related parameters were determined when the leaves reached harvestable size (70 days after sowing). Specifically, vitamin C content was measured by the iodine titration method; reducing sugar content was determined using the DNS method [20]; total organic acid content was analyzed using the Emarcov method [21]; anthocyanin content was determined by the pH differential method [22]; and nitrate content was quantified using spectrophotometry [23].

Microsoft Excel 2010 and SPSS 16.0 were used for statistical analysis via one-way ANOVA (Tukey's test) at α = 0.05.

3 Results and discussion

3.1 Some parameters related to the growth of purple kale

Plant height

Purple kale is a leafy vegetable, where the leaves are harvested gradually from the bottom to the top. Therefore, plant height, along with leaf number and other agronomic traits, is a crucial parameter for evaluating yield potential. In this study, plant height was measured at three time points after transplanting the seedlings into nutrient solutions with varying nitrogen concentrations, as presented in Table 1.

Table 1. Plant height of purple kale (cm) at different growth stages under various nutrient solution treatments

Time after sowing (days)	Nutrient solution treatments						
	Control	T1	T2	T3	T4	T5	T6
30	13.70^{cd} ± 0.62	14.80^{c} ± 0.67	15.10^{b} ± 0.59	16.55^{a} ± 0.64	16.98^{a} ± 1.69	13.28^{d} ± 0.36	4.05^{e} ± 0.24
50	25.13^{b} ± 1.25	28.75^{a} ± 1.51	29.13^{a} ± 1.35	29.58^{a} ± 1.43	29.68^{a} ± 2.11	26.80^{b} ± 0.11	0
70	29.68^{c} ± 1.69	29.90^{c} ± 1.38	31.60^{bc} ± 1.95	32.84^{b} ± 1.93	36.70^{a} ± 2.41	27.84^{d} ± 0.59	0

Note. Different letters in each row indicate significant differences among varieties at $\alpha \leq 0.05$.

The results in Table 1 indicate that plant height generally increased over time across most treatments, except for treatment T6. In this treatment, the nitrogen concentration was excessively high (683.2 mg/L), which led to the development of small, yellow leaves, weak stems, and significantly reduced growth shortly after transplanting. By 30 days after planting, plant height in T6 was only 4.05 cm—the lowest among all treatments—and the plants subsequently died.

Among the remaining treatments, plant height at each time point increased progressively from the control to treatment T4, followed by a slight decline in treatment T5. Specifically, at 30 days after planting, plants in T4 reached the highest height of 16.98 cm, increasing to 29.68 cm at 50 days and 36.70 cm at 70 days. These values were 3.28 cm, 4.55 cm, and 7.02 cm greater, respectively, than those in the control treatment. Similarly, plant heights in treatments T1, T2, and T3 were also greater than in the control by 0.22–3.16 cm at 70 days.

These findings suggest that increasing nitrogen concentration from 170.8 mg/L (Control) to 512.4 mg/L (T4) positively influenced plant height in purple kale. However, when nitrogen levels exceeded this optimum threshold (597.8 mg/L in T5 and especially 683.2 mg/L in T6), plant growth was inhibited, eventually leading to plant death in T6. Similar trends have been reported in the *Persicaria minor*, where an increase in nitrogen concentration from 100 mg/L to 200 mg/L resulted in a 4 cm reduction in plant height [12].

Number of leaves

Nitrogen is known to stimulate the initiation of new leaves from the shoot apical meristem, thereby contributing to an increase in leaf number and, consequently, enhancing the biological yield of plants [6]. An increase in nitrogen levels in the nutrient solution from 5% to 200% nearly doubled the number of leaves in Chinese cabbage [24]. Similarly, in lettuce, the number of leaves increased from 10 to 20 per plant as nitrogen concentration rose from 150 to 250 mg/L, but declined to 14 leaves at 300 mg/L [13]. A comparable trend was observed in our study.

Table 2. Number of leaves per plant at different growth stages under various nutrient solution treatments

Time after sowing (days)	Nutrient solution treatments						
	Control	T1	T2	T3	T4	T5	T6
30	$6.33^{c} \pm 0.32$	$6.48^{c} \pm 055$	$6.80^{b} \pm 0.34$	$7.17^{ab} \pm 0.25$	$7.43^{a} \pm 0.31$	$6.56^{b} \pm 0.27$	$4.50^{d} \pm 0.18$
50	$9.17^{bc} \pm 0.75$	$9.51^{b} \pm 0.49$	$9.83^{ab} \pm 0.41$	$10.50^{a} \pm 0.55$	$10.92^{a} \pm 0.34$	$9.07^{c} \pm 0.40$	0
70	$10.15^{c} \pm 0.58$	$10.24^{c} \pm 0.64$	$11.29^{bc} \pm 0.46$	$12.67^{b} \pm 0.46$	$13.60^{a} \pm 0.37$	$10.32^{c} \pm 0.44$	0

Note. Different letters in each row indicate significant differences among varieties at $\alpha \leq 0.05$.

Specifically, the data presented in Table 2 show that the number of leaves per plant increased progressively as nitrogen concentration rose from 170.8 mg/L (control) to 512.4 mg/L (T4). However, similar to the trend observed in plant height, when nitrogen levels exceeded the optimal threshold, the number of leaves declined in treatments T5 and T6. At 30 days after planting, the average number of leaves per plant increased from 6.33 in the control to a peak of 7.43 in T4, but declined to 6.56 in T5. This trend was further reinforced at 50 days, with T4 reaching the highest leaf count of 10.92 leaves per plant, compared to 9.07 leaves in T5. By 70 days, the leaf number was 13.60 in T4 and 10.32 in T5.

Statistical analysis in Table 2 also indicates no significant differences in leaf number among the control, T2, or between T2 and T3 across all three time points. These results suggest that a nitrogen concentration of 512.4 mg/L (T4) may be optimal for promoting leaf development in purple kale under the experimental conditions.

pISSN 1859-1388
eISSN 2615-9678

Fresh weight per plant.

Variations in plant height and leaf number were accompanied by corresponding changes in the fresh weight of purple kale across the experimental treatments, as presented in Table 3. The data show that the fresh weight increased rapidly after the 30-day period and differed significantly among treatments. Specifically, at 30 days after sowing, fresh weight ranged from 2.22 to 5.78 g/plant, except for treatment T6, where plants recorded a markedly low weight of only 0.51 g/plant.

Table 3. Fresh weight of purple kale plant (g) at different growth stages under various nutrient solution treatments

Time after sowing (days)	Nutrient solution treatments						
	Control	T1	T2	T3	T4	T5	T6
30	$2.22^{d} \pm 0.15$	$3.34^{cb} \pm 0.11$	$3.52^{b} \pm 0.39$	$3.81^{b} \pm 0.37$	$5.78^{a} \pm 0.38$	$3.05^{c} \pm 0.25$	$0.51^{e} \pm 0.09$
50	$20.93^{d} \pm 0.83$	$21.88^{c} \pm 1.64$	$22.05^{c} \pm 0.62$	$24.27^{b} \pm 0.29$	$26.42^{a} \pm 0.27$	$21.13^{c} \pm 0.41$	0
70	$35.34^{e} \pm 1.54$	$46.21^{d} \pm 2.39$	$60.18^{c} \pm 2.87$	$66.05^{ab} \pm 2.27$	$68.93^{a} \pm 2.34$	$27.53^{f} \pm 1.83$	0

Note. Different letters in each row indicate significant differences among varieties at $\alpha \leq 0.05$.

As the root systems developed and adapted to the nutrient-rich hydroponic environment—where minerals are readily available in dissolved form—nutrient uptake was enhanced, leading to vigorous plant growth and a sharp increase in plant height, leaf number, and consequently, fresh weight. From day 30 to day 70, the fresh weight of purple kale increased more than tenfold compared to the earlier stage. The highest fresh weight was recorded in treatment T4, reaching 68.93 g/plant. In comparison, T3 and T2 yielded 66.05 g/plant and 60.18 g/plant, respectively—approximately 15–20 g/plant higher than T1 (46.21 g/plant) and nearly double that of the control (35.34 g/plant).

In contrast, growth inhibition due to excessive nitrogen resulted in a notable reduction in fresh weight in T5. This treatment yielded only 27.53 g/plant—approximately 40% of the fresh weight in T4—and lower than all other experimental treatments and the control.

These findings indicate that increasing nitrogen concentrations in the Knop solution significantly enhanced the growth performance of purple kale, as evidenced by improvements in plant height, leaf number, and fresh weight. Among the treatments, the formulation with a threefold increase in nitrogen (T4) yielded the best results for all growth parameters, followed by T3 and T2. A 1.5-fold increase in nitrogen (T1) resulted in moderate improvement compared to the control, while a 3.5-fold increase (T5) tended to suppress growth. At the highest nitrogen concentration (T6, fourfold the original Knop level), plant growth ceased, and all plants died within approximately 30 days of hydroponic cultivation.

The observed toxicity symptoms may be attributed to ion uptake imbalances, particularly involving nitrate (NO_3^-) interactions with other ions in the nutrient solution. Excessive NO_3^- uptake has been shown to significantly reduce Fe accumulation in leaves, leading to iron deficiency symptoms—such as chlorosis and growth inhibition—as demonstrated in various studies [25, 26]. This explains the yellowing leaves, weak stems, underdeveloped roots, and eventual plant death observed in T6. Moreover, excessive NO_3^- uptake may also impair the absorption of other essential ions, such as Cl^-, SO_4^{2-}, and PO_4^{3-}, thereby causing nutrient imbalances and growth suppression in plants exposed to high nitrogen concentrations [26]. In addition, nitrogen toxicity observed in T6 may also be related to salt stress.

Total chlorophyll content

Nitrogen plays a crucial role in photosynthesis as it is a key component of the photosynthetic apparatus, contributes to increasing leaf area, delays chlorophyll degradation, and affects leaf longevity [27, 28]. The effects of nitrogen concentration on the chlorophyll content in purple kale leaves are shown in Table 4.

According to Table 4, total chlorophyll content increased progressively from day 30 to day 70 in all treatments except for treatment T6. An increase in nitrogen concentration from the control up to T4 was associated with higher pigment accumulation. Specifically, at all three time points, the highest chlorophyll content was recorded in treatment T4 (512.4 mg N/L), reaching 2.12, 2.23, and 2.71 mg/dm², respectively—1.2 to 1.7 times higher than that of the control at the same stages.

Treatments T1, T2, and T3 showed slightly higher chlorophyll content than the control, but no statistically significant differences were observed among these treatments at 30 and 50 days. However, at 70 days, chlorophyll content in T3 was statistically similar to that in T4. When nitrogen concentration exceeded the optimal threshold, chlorophyll content tended to decline. At day 30, the lowest chlorophyll content was recorded in treatment T6 (0.89 mg/dm²), which was only 75.42% of the control. Experimental observations revealed small, yellowish leaves in this treatment, which could be associated with iron deficiency symptoms induced by excessive nitrate concentrations in the nutrient solution, as previously discussed [25]. At 50 and 70 days, the lowest chlorophyll content was found in treatment T5, at 1.42 and 2.15 mg/dm², respectively.

Table 4. Total chlorophyll content (mg/dm²) of purple kale at different growth stages under various nutrient solution treatments

Time after sowing (days)	Nutrient solution treatments						
	Control	T1	T2	T3	T4	T5	T6
30	$1.18^{c} \pm 0.08$	$1.45^{bc} \pm 0.20$	$1.53^{b} \pm 0.09$	$1.55^{b} \pm 0.04$	$2.02^{a} \pm 0.16$	$1.34^{bc} \pm 0.06$	$0.89^{d} \pm 0.07$
50	$1.61^{b} \pm 0.05$	$1.62^{b} \pm 0.14$	$1.64^{b} \pm 0.13$	$1.68^{b} \pm 0.21$	$2.33^{a} \pm 0.15$	$1.42^{c} \pm 0.09$	0
70	$2.21^{bc} \pm 0.13$	$2.32^{b} \pm 0.17$	$2.35^{b} \pm 0.21$	$2.65^{a} \pm 0.32$	$2.71^{a} \pm 0.10$	$2.15^{c} \pm 0.29$	0

Note. Different letters in each row indicate significant differences among varieties at $\alpha \leq 0.05$.

A similar trend was reported in *Persicaria minor*, where total chlorophyll content increased with nitrogen levels from 0 to 200 mg/L [12]. Likewise, in Buttercrunch lettuce, increasing nitrogen concentration from 150 to 300 mg/L in hydroponic solution led to higher chlorophyll content, although in the Black Seeded Simpson variety, no increase was observed when nitrogen increased from 250 to 300 mg/L [13]. The strong correlation between leaf nitrogen content and chlorophyll concentration is well-established, as up to 70% of the nitrogen in leaves is localized in chloroplasts, where chlorophyll is synthesized [29]. Sufficient nitrogen supply has also been reported to enhance chlorophyll content in wheat leaves [30].

The results of the present study also support this trend: increasing nitrogen

pISSN 1859-1388
eISSN 2615-9678

concentration from 170.8 mg/L to 512.4 mg/L resulted in a marked increase in total chlorophyll content in purple kale leaves. This improvement in pigment content positively influenced other growth parameters, including plant height, leaf number, and fresh weight.

3.2 Some parameters related to the quality of purple kale

Total organic acid, vitamin C, and reducing sugar content

To evaluate the effects of nitrogen on the quality of purple kale, the study analyzed several quality-related parameters, with results summarized in Table 5. The analysis showed significant differences among the experimental treatments and the control across all three quality indicators (Table 5). Specifically, increasing the nitrogen concentration in the nutrient solution from 170.8 mg/L (control) to 683.2 mg/L (T5) led to an increase in both vitamin C content and reducing sugars in kale leaves, while the total organic acid content decreased in treatment T5.

Among the treatments, T2 (341.8 mg N/L) produced the most favorable results across all three indicators. Plants grown in T2 accumulated 55.61 mg/g of total organic acid—1.97 times higher than the control. Vitamin C content in T2 reached 58.66 mg/100g, which was 1.69 times higher than that of the control. Reducing sugar levels showed no significant differences among T2, T3, and T4, with values ranging from 4.46 to 4.62 mg/g—roughly twice the reducing sugar content recorded in the control treatment.

The reducing sugar content in purple kale observed in this study was higher than that of green kale (3.5–3.8 mg/g) grown in Grow Master and BKFast nutrient solutions [17]. However, the vitamin C content of purple kale was only about 72.48% of that found in green kale in the same study and approximately 92.2% of the value reported in [31].

Table 5. Total organic acid, vitamin C and reducing sugar content of purple kale at different growth stages under various nutrient solution treatments

Traits	Nutrient solution treatments					
	Control	T1	T2	T3	T4	T5
Total organic acid content (mg/g)	$28.13^{d} \pm 1.75$	$35.54^{c} \pm 2.24$	$55.61^{a} \pm 4.02$	$43.03^{b} \pm 2.39$	$28.43^{d} \pm 1.35$	$21.70^{e} \pm 1.46$
Vitamin C content (mg/100g)	$34.64^{e} \pm 1.61$	$44.79^{b} \pm 3.43$	$58.66^{a} \pm 3.49$	$43.71^{c} \pm 3.56$	$37.35^{d} \pm 2.77$	$35.40^{de} \pm 1.85$
Reducing sugar content (mg/g)	$2.25^{d} \pm 0.12$	$3.37^{c} \pm 0.23$	$4.46^{a} \pm 0.17$	$4.58^{a} \pm 0.36$	$4.62^{a} \pm 0.27$	$3.98^{b} \pm 0.29$

Note. Different letters in each row indicate significant differences among varieties at $\alpha \leq 0.05$.

Anthocyanin, and nitrate content

Anthocyanin content

Anthocyanins are known for their vibrant pigmentation and play a crucial role as potent antioxidants. These natural flavonoid pigments are responsible for red, purple, and blue colors in plants. The anthocyanin content in purple kale can vary depending on the cultivar and growing conditions, especially temperature, light, and nutrient availability [32, 33]. The analysis of anthocyanin content in purple kale leaves at 70 days after transplanting is presented in Table 6.

Statistical analysis revealed no significant differences in anthocyanin content between treatments T1 and T2 compared to the control, with values ranging from 0.61 to 0.68 mg/g fresh weight (Table 6). However, increasing nitrogen concentrations in the nutrient solution from T2 to T5 (341.8 to 683.2 mg N/L) resulted in a gradual decline in anthocyanin accumulation compared to the control and T1. Specifically, anthocyanin contents in T3, T4, and T5 were only 85.2%, 67.2%, and 60.6% of the control, respectively, with the lowest value being 0.37 mg/g in T5. Previous studies have reported that nutrient deficiency, particularly nitrogen deficiency, enhances anthocyanin accumulation in red cabbage and red kale [34]. Similarly, increasing nitrogen levels from 0 to 270 kg/ha progressively reduced anthocyanin content in leaves, stems, and roots of Labisia pumila, a medicinal herb [35].

Table 6. Anthocyanin and nitrate content of purple kale at different growth stages under various nutrient solution treatments

Traits	Nutrient solution treatments					
	Control	T1	T2	T3	T4	T5
Anthocyanin content (mg/g)	$0.61^{a} \pm 0.07$	$0.68^{a} \pm 0.05$	$0.65^{a} \pm 0.10$	$0.52^{c} \pm 0.06$	$0.41^{d} \pm 0,07$	$0.37^{e} \pm 0.09$
Nitrate content (mg/kg)	$577.86^{e} \pm 14.60$	$763.74^{d} \pm 15.41$	$785.54^{d} \pm 12.15$	$889.25^{c} \pm 11.60$	$921.45^{b} \pm 16.20$	$1145.67^{a} \pm 15.92$

Note. Different letters in each row indicate significant differences among varieties at $\alpha \leq 0.05$.

This decrease is consistent with previous reports in other plant species, where limited nitrogen supply has been shown to enhance anthocyanin accumulation by promoting the expression of key structural and regulatory genes involved in the phenylpropanoid biosynthetic pathway [36]. The relatively high ambient temperatures during the experiment (22–27°C) likely contributed to the low anthocyanin levels observed, as cooler conditions have been shown to promote anthocyanin accumulation through activation of regulatory genes [31]. This may explain why anthocyanin content in purple kale in this study was markedly lower than that reported for the Red Dove cultivar (1.73 mg/g) [31].

Nitrate content

The results presented in Table 6 clearly demonstrate the influence of nitrogen concentration in the nutrient solution on nitrate accumulation in purple kale leaves at harvest. Specifically, nitrate content increased linearly with the rise in nitrogen concentration supplied through the nutrient solution. The control treatment, with the lowest nitrogen level, showed an average nitrate content of 577.86 mg/kg. As the nitrogen concentration increased across treatments T1 to T5, nitrate content in the leaves also increased correspondingly, reaching a maximum value of 1145.67 mg/kg in T5.

The increase in nitrate accumulation with higher nitrogen supply can be attributed to the plant's nitrogen uptake and assimilation mechanisms. Nitrogen is an essential nutrient that plays a critical role in numerous biochemical and physiological processes, including the synthesis of proteins, nucleic acids, and chlorophyll. When nitrogen is abundantly available, plants tend to

pISSN 1859-1388
eISSN 2615-9678

absorb large quantities of it in the form of nitrate (NO_3^-) or ammonium (NH_4^+). However, when nitrogen supply exceeds the plant's immediate needs for growth and development, the processes of nitrate reduction to nitrite and subsequently to ammonium—necessary for incorporation into organic compounds—may not keep pace with uptake. This imbalance leads to the accumulation of nitrate in plant tissues, particularly in the leaves [37].

Similar trends in nitrate accumulation have been observed in other leafy vegetables [38]. For instance, in Chinese cabbage, nitrate content under a 200% nitrogen treatment reached 1300 mg/kg - 1.86 times and over 13 times higher than under treatments with 100% and 5% nitrogen, respectively [23].

The accumulation of nitrate in leafy vegetables is an important factor that requires attention due to its potential health risks associated with the consumption of foods containing high nitrate levels. Studies have shown that nitrate can be reduced to nitrite in the human body, and nitrite may cause health issues such as methemoglobinemia, particularly in infants and young children [39]. Therefore, controlling nitrate levels in vegetables, including purple kale, is a crucial requirement for safe agricultural production.

The maximum allowable nitrate content (mg/kg fresh weight) varies across different countries and regions depending on the type of vegetable. According to the European Commission, the maximum permissible nitrate levels in leafy vegetables are regulated under Commission Regulation (EC) No 1258/2011, depending on the type of vegetable and the season. Specifically, the limits for certain vegetables such as lettuce and spinach range from 2,000 to 4,500 mg/kg fresh weight [39, 40]. The World Health Organization (WHO) recommends an acceptable daily intake (ADI) of nitrate at 3.7 mg per kg body weight, equivalent to approximately 222 mg for a 60-kg adult. Thus, the nitrate levels measured in purple kale in this study remain well within these safety limits, indicating no immediate health risk when consumed in typical amounts.

The summary of growth and quality traits of purple kale under different nitrogen concentrations is presented in Table 7.

The results indicate that the T4 treatment (512.4 mg N/L) had the most favorable effect on plant growth, as evidenced by its lowest total score for growth-related parameters, including plant height, leaf number, fresh weight, and chlorophyll content. In contrast, T2 (341.8 mg N/L) was most effective in enhancing quality attributes such as organic acid, vitamin C, reducing sugar, and anthocyanin contents, reflected by its lowest total score for quality traits. These findings suggest that optimal nitrogen concentration for maximizing vegetative growth may differ from that required to improve crop quality. Similar trends have been reported in leafy vegetables, where high nitrogen levels promote biomass accumulation, whereas moderate levels favor the synthesis of secondary metabolites and antioxidants [41]. This pattern has also been observed in other crops such as sweet potato and *Persicaria minor* [11, 12]. Therefore, the selection of nutrient regimes should consider the specific production goals—either for biomass yield or nutritional quality.

Table 7. Ranking of nitrogen treatments based on growth and quality parameters of purple kale

Traits	Nutrient solution treatments						
	Control	T1	T2	T3	T4	T5	T6
Some parameters related to the growth of purple kale							
Plant height	4	4	3	2	1	5	6
Number of leaves per plant	4	4	3	2	1	4	5
Fresh weight per plant	5	4	3	2	1	6	7
Total chlorophyll content	3	2	2	1	1	4	5
Total scores	**16**	**14**	**11**	**7**	**4**	**19**	**23**
Some parameters related to the quality of purple kale							
Total organic acid content	4	3	1	2	4	5	-
Vitamin C content	6	2	1	3	4	5	-
Reducing sugar content	4	3	1	1	1	2	-
Anthocyanin content	1	1	1	2	3	4	-
Nitrate content	1	2	2	3	4	5	-
Total scores	**16**	**11**	**6**	**11**	**16**	**21**	**-**

Note. Traits were ranked from 1 (most favorable) to 7 (least favorable) based on a scoring system derived from experimental results. Total scores were calculated separately for growth and quality groups. Lower total scores indicate greater positive effects. "–" indicates data not assessed.

4 Conclusion

Nitrogen concentration of 512.4 mg/L maximized growth of purple kale, whereas excessive supply (≥597.8 mg/L) suppressed development and caused mortality at 683.2 mg/L. For quality attributes, 341.8 mg/L produced the highest organic acids, vitamin C, reducing sugars, and anthocyanins, while maintaining nitrate within EU safety limits. Thus, 341.8 mg/L is recommended for production targeting nutritional quality, with EC monitoring to optimize nutrient use in commercial systems. Future studies should explore the use of organic fertilizers and real-time nitrogen monitoring with IoT technologies to further enhance sustainability and precision in leafy vegetable production.

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pISSN 1859-1388
eISSN 2615-9678

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