



Research Article

Effect of Spraying with Conventional and Nano-NPK Fertilizer on the Growth and Chemical Content of Lettuce Plants Grown by Hydroponic Technique

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Abstract | The current study was conducted during the winter season 2022-2023 in a hydroponic system in Al-Diwaniyah Governorate, Iraq, to investigate the impact of spraying with conventional and nano-NPK fertilizer on several vegetative Growing properties and composition of *Lactuca sativa var. capitata L.*, the experiment included two fertilizer treatments, 2 and 4 g/l of conventional and nano-NPK fertilizer, in addition to an un-fertilized control group. The results showed significant superiority of Nano NPK 4g/l treatment in the characteristic of leaf number, vitamin K, phosphorus, sulfur and zinc with values of 28 leaves/plant, 149.77 mg/100 g fresh weight, 0.27%, 31.27 mg/g dry weight, 3 mg/100 g fresh weight respectively compared to plants treated with Nano NPK 2g/l and control group which gave the lowest values. Whereas, Nano NPK 2g/l treatment was significantly superior in wet weight, dry weight, leaf area, carbohydrates, vitamins A and C, potassium, iron, calcium and fiber with values of g 160, 11.5 g, 13150.05 cm²/plant, 1.49%, 204.32 mg/100 g fresh weight, 4.48 mg/100 g fresh weight, 1.3%, 2.35%, 57.4 mg/100 gm, 1.85% respectively compared to plants treated with Nano NPK 4g/l and control group which gave the lowest values. In addition, conventional NPK showed superiority in plant height, total chlorophyll, carotenoids, protein and nitrogen among all treatments with values reaching 27.33 cm, 4.61 mg/gm fresh weight, 0.21 mg/gm, 1.79% and 2.36%. Statistical analysis confirmed considerable variations were found between several tests at $p \leq 0.05$.

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Introduction

Nutritional balance is essential to obtain a high yield with good quality, so added fertilizers are of great importance, which are added by spraying or adding to the soil Al-Kaisy and Al-Mgadami (2014). Nitrogen is responsible for the synthesis of

essential substances like NAD, NADP, nitrogenous bases, proteins, amino acids, protoplasm, chlorophyll, growth stimulants, and other substances required for the building of plant cells, as well as phosphorus, which is involved in essential components including RNA, DNA, phosphorylated lipids, NADP and NADP enzymatic chaperones, energy compounds

ATP, GTP, and can be useful in the generation of vital substances including protein and is involved in carbohydrate metabolism, Potassium is a critical plant nutrient involved in numerous physiological processes. It regulates cell membrane function, controls stomata opening and closing, activates essential enzymes, mitigates calcium toxicity, and influences the synthesis of carbohydrates, lipids, and proteins. Potassium also plays a pivotal role in energy metabolism (Fathi, 2022; Mostofa *et al.*, 2022; Nicholls *et al.*, 2023).

Among the most important issues faced by farmers is the deterioration of the soil over time, as its natural composition declines and salts accumulate, in addition to the spread of diseases and pests (Shahane and Shivay, 2021). For these reasons, researchers in the agricultural sciences are looking for alternative solutions to use of soil, including soilless cultivation techniques or hydroponics, in which plants are grown in media other than soil, which depends mainly on the kind of nutritive solution and the media provides stabilization and assistance to the plant. The formulation of nutrient media differs in ingredients and ratios for various plants, particularly for lettuce, based on weather conditions and phase of growth (Mir *et al.*, 2022).

Lettuce (*Lactuca sativa* L.), belonging to the Asteraceae family, is a major winter leafy veggie in Iraq, native to the Mediterranean region, Europe, Canada, USA, Latin America and Northern Africa and is considered one of the world's most important leafy greens, sought after by consumers for its rich nutritional properties Krüger (2020).

Lettuce contains several important minerals, including iron, phosphorus, calcium, iodine, chlorine, magnesium, cobalt, and carotene. Lettuce is also used as a moisturizer, pain reliever, blood cleanser, and vision booster because it contains vitamin A (Mou, 2009). Lettuce is an amazing resource of health-promoting natural constituents such as polyphenols, carotenoids and chlorophyll. However, the nutritional and antioxidant profile varies between lettuce varieties, particularly between green and red. The advantages of consuming lettuce are based on its content, especially the presence of antioxidants that can act as a nutraceutical (Shi *et al.*, 2022).

Nano-fertilizers can meet the nutritional needs of plants and guarantee the efficiency and durability

of agricultural productivity systems while ensuring that plant performance is not compromised. Nanotechnologies use nanosized materials in the range of 1-100 nanometers; in this size range, particles have specific characteristics that allow an effective interaction at the intended location (Kumar *et al.*, 2021). The nanoscale form of specific trace elements allows for better plant absorption of micronutrients and thus improves plant development and nutritive value (Elemike *et al.*, 2019). Nanofertilizers can be delivered in either a solid or sprayed form, which improves the deliver capability of minerals to the plant and increases the uptake capacity of traditional inorganic fertilizers. A few of the shared features of Nanofertilizers are (1) provision adequate nutrition through leaf and soil amendments to promote plant development, (2) Environmentally efficient and economical, (3) their very efficient fertilization (4) their complementary function to inorganic fertilizers, (5) protect the surrounding area from pollutants risks along with the elimination of contamination of water (Jakhar *et al.*, 2022).

Helaly *et al.* (2021) indicated that treatment lettuce plants with nano NPK markedly boosted plant development in terms of fresh plant weight, leaf area, fresh head weight, head diameter, strength, gross weight and merchantable value. Also, another study improved considerably enhanced in most of the plant development characteristics and total income content of lettuce plants treated with foliar application of nano NPK at (4 g/l) Sulaiman and Rasheed (2024).

The study aimed to assess the impact of spray usage of conventional and nano-NPK fertilizer on the vegetative and chemical contents of hydroponically grown lettuce plants.

Materials and Methods

Experimental design and procedure

This experiment used a completely randomized design (CRD) for a factorial experiment with two factors and three replicates (control group sprayed with distilled water, Nano-NPK 2g/l, Nano-NPK 4g/l, NPK 2g/l, 4g/l).

Hydroponics technique

The procedure was performed in a domestic setting in Diwaniya Governorate, Iraq. The hydroponic system consisted of 5 iron stands arranged at a distance of

50 cm between one stand and another and welded vertically with the roof and base to increase its support and bear the weight of the system, each stand carries two pipes 4 m long and 4 inches in diameter and the distance between the height of one pipe and another is 40 cm, and the system included two water tanks with deionized water RO (Reverse Osmosis). The tanks have a water plunger connected to a water hose to patch the water from the tanks to the pipes using NFT membrane technology and supply it to the plants during this time.

Planting seeds and transporting seedlings

In the 2022-2023 growing season, *Lactuca sativa* var. *capitata* lettuce seeds produced by the Turkish company Arzuman, which can be grown using NFT hydroponics, were planted in 40 well plastic trays with 10 trays each using Dutch Patmos as the growing medium inside Al-Rahman Al-Nursery and Gardens in Al-Arouba district, Al-Diwaniyah governorate, on 03/10/2022. At the rate of watering every day and then based on the needs of the plant for water as the plants progressed in growth and entered the winter season, the first appearance of the seedlings after 5 days, then the saplings were moved to the plantation after the age of (4-5) leaves, and after 10 days of seedling stabilization, the nutrient solutions were pumped in addition to Ca spraying every 15 days.

The date of the first fertilizer treatment was 23/11/2022. After foliar spraying with a one-liter hand sprayer at the known concentration mentioned above for each treatment, the spraying process was repeated on 03/12/2022.

Characteristics examined

Vegetative growth indicators: Plant height (cm):

In each transect and for all replicates, the plant height was determined by the surface of the cup in the planter to the top of the plant using a ruler, and the average has been estimated by dividing the total height of the plants in any treatment by the number of plants [Singh and Stoskopf \(1971\)](#).

Leaves number (leaves/plant)

The leaves number for every replicate in each treatment was calculated, then the average was derived by division of the total leaves number in each treatment by the plant number in that treatment.

Fresh and dry weight (g/plant)

Each test with three plants was randomly selected and weighed using a sensitive electronic balance, and the average was derived from the sum of the weights of the plants in each treatment divided through plant numbers, the vegetative sum of each plant was dried at room temperature and airflow until the weight was stable, and the average dry weight was derived from the sum of the plant dry weights of the in each test divided by the plant number.

Total leaf area (cm²/ Plant)

Leaf area was measured using the method adopted by Al-Zaidy and Al-Ubaidy 2017 by taking three full-width leaves from each replicate using a scanner and image software installed on a laptop. The average area of a leaf was then multiplied by the average number of lettuce leaves per replicate to calculate the plants' total leaf area.

Chemical content indicators

Total chlorophyll (mg/g.f.wt): The amount of chlorophyll in the leaves was estimated according to the method of [Mackinney \(1941\)](#), one g of fresh leaves was weighed, assorted well with 10 ml of 80% acetone in a glass test tube and left in the refrigerator at 4°C in the dark for 20 hours, then shaken again and left in the same conditions for 1-2 hours, then centrifugated with 3000 rpm for 10 min., the filtrate was acidified with 2-1 drops of hydrochloric acid (0.1 N), the filtrate was diluted twice with acetone, and the total chlorophyll content of the leaves was calculated via a wavelength spectrophotometer (645 and 663 nm) according to the following equation:

$$Total\ Chlorophyll\ (mg/g\ tissue) = [20.2(D_{645}) + 8.02(D_{663})] \times V / (1000 \times W)$$

Where; D = optical density reading of the chlorophyll extract at the specific. Wavelength, V = final volume of the 80% acetone-chlorophyll extract. W = fresh weight in grams of extracted tissue.

Estimation of carotenoids (mg/g)

Carotenoids were estimated by the method described in [Kirk and Allen \(1965\)](#) using a spectrophotometer for carotenoid estimation. at 480, 663 and 645 nm.

$$Carotenoid\ (mg/g) = \frac{[A_{480} + 0.114A_{663}] - 0.638A_{645}}{A \times 1000 \times W} \times V$$

V = Final filter volume (ml), A = Optical density of

chlorophyll extracted in the cell (1 cm), W = Leaf fresh weight (g).

Estimation of total carbohydrates (%)

Carbohydrates were estimated by the modified phenol-Sulfuric acid colourimetric technique described by DuBois *et al.* (1956), the method is widely used because of its sensitivity and simplicity, The amount of total dissolved carbohydrates is calculated according to the following equation:

$$\text{Total sugar concentration} = \frac{A_{\text{test}}}{A_{\text{STD}}} * \text{Conc. of STD}$$

Protein estimation (%)

The protein concentration in both crude and purified extracts was determined using the Bradford (1976), which involves dissolving Coomassie Brilliant Blue G-250 dye in 100 ml of 85% phosphoric acid and 50 ml of ethyl alcohol. The volume was then made up to 1 liter, filtered through filter paper and the standard curve of bovine serum albumin concentration ($\mu\text{g/ml}$) was plotted against absorbance at a wavelength of 595 nm.

Vitamin A (mg/100 g. f. wt.)

Vitamin A can be valued according to the procedure of Jadoon *et al.* (2013) using a ferrozine-ferrous complex, which can be prepared in water and is stable in acetate buffer at pH (6) The ferrous ferrozine complex formed was measured at an absorbance of 562 nm.

Vitamin K (mg/100 g. f. wt.)

Vitamin K was estimated by the method Hassan 1980. 5 g of fresh plant tissue was collected and homogenized with hexane. The spectrophotometric determination of vitamin K depends on the reaction of vitamin K with alcoholic sodium hydroxide in alkaline solutions with concentrations not exceeding 0.2 M% ($p \geq 0.05$).

Estimation of total vitamin C (ascorbic acid) (mg/100 g. f. wt.)

The amount of vitamin C in lettuce was estimated using the 2,4-dinitrophenylhydrazine (DNP) procedure set out in Al-Ani *et al.* (2007). This method is widely used to determine the amount of ascorbic acid in biological fluids. Oxidation of ascorbic acid to dehydroascorbic acid by copper ions and then to 3-diketo-gulonic acid, which interacts with 2,4-dinitrophenylhydrazine to

create red dihydrazone, which is detected at a 520 nm wavelength.

Estimation of total nitrogen (%) by spectrophotometry

This technique is based on Temminghoff and Houba (2004). The Berthelot reaction is employed for the quantification of nitrogen, which is based on the composition of an azo dye in the addition of ammonia and a hypochlorous reagent in an alkaline solution; indophenols with nitrogen have a bluish-green color, and the absorption is being read at a 660 nm wavelength. This is a measurement of the ammonium values and this technique is effective in measurement of the content of nitrogen compounds in the test sample.

Leaves phosphorus content (%)

The phosphorus concentration in the leaves was assessed using the ammonium molybdate and ascorbic acid method Bender and Wood (2000). 10 ml of the digested sample was taken and placed into a 50 ml graduated flask, and the contents were made up to volume using deionized water, then 10 ml of the previous solution was recorded and placed in a 100 ml Erlenmeyer flask and 0.1 g of ascorbic acid and add 4 ml of ammonium molybdate. 1 g ascorbic acid and 4 ml ammonium molybdate were added and the sample exposed to heat via hot plate for one minute until the solution turned blue, the contents of the flask were then transferred quantitatively to a 100 ml graduated flask and make up to the level with deionized water, the reading was recorded at a wavelength of 620 nm in the spectrophotometer and optical absorbance readings were taken for a series of phosphorus standard concentrations solutions to construct a phosphorus standard curve by drying 2.5 g potassium phosphate. 5 g of KH_2PO_4 was dried in the oven at 105 °C for 1 h. After cooling in the oven, 0.439 g was removed and dissolved in 1 liter of deionized water (to give 100 mg phosphorus). Standard solutions were prepared by taking 0, 0.5, 1, 1.5, 2 and 2.5 ml, the optical densities of these solutions were then measured, the standard curve was plotted and the units of measurement for the values obtained were converted to units of concentration as %.

Leaf content of potassium (%) and calcium (mg/100 g)

The procedure was carried out with the application a flame photometer approved by Tomar and Agarwal (2013). 0.5 g of dehydrated plant tissue was collected in an Erlenmeyer flask and disrupted in a triple acid

mix ($\text{HNO}_3 + \text{HClO}_4 + \text{H}_2\text{SO}_4$) in the ratio of (3:1:9). the discolored decomposition residue was separated by filter Whatman No. 1 into a 100 ml graduated flask to decrease whole volume to 25 ml. A 5 ml portion was aliquoted and filled up to 25 ml with deionized water and measured immediately on a digital flame-photometer using separate K and Ca filters and the percent K and Ca were plotted against calibration curves.

Zinc determination method (mg/100 g. f. wt.)

The Abnova complete kit was used to measure the amount of zinc (Zn) after the appropriate digestion of the sample. According to [Knoell et al. \(2009\)](#). The Zinc test is a kit developed to detect zinc in direct measurement in biomaterial sources without any pre-treatment. The current procedure uses a chromogen that forms a color when it binds with zinc. The color intensity detected at 425 nm, is immediately related to the zinc values in the sample.

Iron (Fe) (%)

The iron content of the floral disc was estimated according to the method of [Sandell \(1951\)](#). Samples of treated plants were taken and drying in an oven at 70°C up to a steady weight, then ground and added to 1 ml of Sulfuric acid (H_2SO_4) and leave to sit for one hour, then incinerated in an oven until the ashes had turned white. for one hour and then incinerated in an oven until the ash was white, then dissolve the ash in 5 ml of HCl 6N. and filtered through an acid-washed filtering disc and makeup to 100 ml with HCl (0.1 N).

Twenty-five ml of the solution was taken and a volume of pre-prepared sodium citrate solution was added until the pH was 3.5, then 1 ml of hydroquinone and 1 ml of Ortho-phenanthroline solution were added and the solution was left at 20°C for one hour to reduce the amount of iron present. Iron standards have been generated at values of 5, 10, 20, 30, 40, 50, 70 and 90 mg/l to plot the iron standard curve and the OD was recorded using a spectrophotometer at 508 nm wavelength.

Sulfur estimation mg/gm dry weight

Sulfur was estimated by the method of [Saxena \(2001\)](#) using three solutions (sodium chloride-hydrochloric acid solution, glycerol-ethanol solution, barium chloride and sulfate solution) by drying 5 ml of plant tissue and digesting it with 50 ml of distilled water to obtain a regular suspension. Filter the suspension

through Whatman filter paper. Take 50 ml of the filtrate and add 10 ml of NaCl-HCl solution, 10 ml of glycerol-ethanol solution and 0,15 g of barium chloride, then mix and stir with a magnetic stirrer for about one hour to ensure proper mixing. The absorbance of the above samples was measured at λ max 420 nm using distilled water as a blank. The calibration curve was then plotted using standard sulphate solutions at different concentrations, the absorbance of each was recorded and the sulfur values were extracted from the calibration curve.

Estimation of crude fiber (%)

Fiber was evaluated using the procedure of [Song et al. \(2018\)](#) by handling lettuce plant samples with heated, diluted Sulfuric acid with a concentration of 12.5% as the initial step (digestion with acid), followed by treatment with heated dilute NaOH with a concentration of 12.5% (digestion with base), then dried and burning the residues and calculating the percentage of fiber by difference.

Statistical analysis

Outcome results were analyzed statistically with SPSS v26 with the two-way analysis of variance test. Values were analyzed by Least Significant Difference (LSD) test at $P \leq 0.05$.

Results and Discussion

The data of [Table 1](#) showed that the highest height of the lettuce plant was 27.33 cm in the case of treatment with NPK 4g/l and the lowest in the control treatment with a height of 18 cm, and the highest leaves number was 28 leaves/plant in the Nano NPK 4g/l treatment while the lowest leaves number was 20.66 leaves/plant in the control treatment. The soft and dry weight recorded the highest value of 160 and 11.5 g in the Nano NPK 2g/l treatment, respectively, and the lowest in the control group, which reached 121 and 10.03 g, respectively. The significant effect of the Nano NPK 2g/l treatment was also observed in the leaf area of the plant, which reached 13150.05 cm^2 relative to control plants, whose leaf area reached 4113.09 cm^2 .

Total chlorophyll and carotenoids recorded the highest values of 4.61 mg/g.f.wt. and 0.21 mg/g in treatment with NPK 4g/l, respectively, and the lowest values were recorded at 1.72 mg/g.f.wt. and 0.13 mg/g in the control treatment, respectively. Total

carbohydrates and proteins recorded the highest values of 1.49% and 1.79% in plants treated with Nano NPK 2g/l and NPK 4g/l, respectively, and the lowest values of 1.03% and 0.97%, respectively, in the control group.

The results indicated that Nano NPK 2g/l treatment significantly enhanced both vitamin A and vitamin C levels with values of (204.32 and 4.48) mg/100g.f.wt respectively. In contrast, vitamin K recorded its highest value (149.77mg/100g.f.wt), respectively. In contrast, the highest vitamin K content (149.77 mg/100g fresh weight) was observed in the Nano NPK 4g/l treatment. The control plants, however, showed the lowest levels of vitamins A and C with values of (133.76 and 2.37 mg/100g.f.wt) respectively, while the lowest value for vitamin K was recorded at 30.8 mg/100g.f.wt in plants treated with NPK 2g/l.

The results of Table 1 displayed that the highest nitrogen and phosphorus contents were 2.36 and

0.27% when treated with NPK 4g/l and Nano NPK 4g/l, respectively, while the lowest values of 1.50 and 0.2% were recorded in the control group for nitrogen and phosphorus, respectively. It was also observed that Nano NPK 2g/l treatment was significantly more effective in plant potassium content, reaching 1.3% when relative to control plants, which had the lowest value of 0.65%.

Based on the results presented in Table 1, the Nano NPK 4g/l treatment had the highest significant effect on S and Zn content with values of (31.27 mg/g dry wt. and 3 mg/100g.f.wt.), respectively, while the control plants had the lowest values of (10.59 mg/g dry wt. and 2.89 mg/100g.f.wt.) for both elements, respectively. Iron, calcium and fiber recorded the highest values of 2.35%, 57.4 mg/100g and 1.85%, respectively, when treated with Nano NPK 2g/l and the lowest values of 1.32%, 29.05 mg/100g and 1.33% were recorded in the control group and NPK 2g/l, respectively.

Table 1: Effect of foliar spraying with conventional and nano NPK fertilizer on vegetative and physiological characteristics of *Lactuca sativa* var. capitata.

Plant parameters	Treatments					LSD
	Control	2g NPK	4g NPK	2g Nano-NPK	4g Nano-NPK	
	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	
Plant height cm	18±1.73205B	24±0.57735A	27.33±2.848A	24.66±0.33333A	19.66±1.66667B	4.39
Number of leaves	20.66±0.88192B	26.33±1.45297A	26.33±0.88192A	25.66±0.88192A	28±3.05505A	4.24
Fresh weight g	121±0.57735D	124±1.73205C	144±1.1547B	160±0.57735A	159±0.57735A	2.64
Dry weight g	10.03±0.09280A	11.2±0.41633A	11±0.28868A	11.5±1.73205A	11±1.1547A	N. S
Total leaf area cm ² /plant	4113.09±0.5773D	6127.63±0.5773A	5362.88±0.5773C	13150.05±0.5773E	5472.45±0.5773B	26.4
Total chlorophyll mg/g.f.wt	1.72±0.05774D	2.73±0.01732C	4.61±0.05774A	3.7±0.02887B	2.06±0.03464C	0.104
Carotenoids mg/g	0.13±0.01732B	0.14±0.00577B	0.21±0.00577A	0.19±0.00577A	0.19±0.00577A	0.023
Carbohydrates %	1.03±0.01732C	1.05±0.02887C	1.07±0.01732C	1.49±0.02887A	1.28±0.02887B	0.066
Protein%	0.97±0.01732D	1.12±0.01155C	1.79±0.00577A	1.35±0.02887B	1.12±0.01155C	0.046
Vitamin A mg/100g.f.wt	133.76±0.88192C	173.90±0.97311B	204.22±1.1547A	204.32±1.2547A	201.06±2.3094A	3.56
Vitamin C mg/100g.f.wt	2.37±0.17321C	2.4±0.00577C	2.79±0.05774C	4.48±0.05774A	3.14±0.11547B	0.8
Vitamin K mg/100g.f.wt	108.47±11.4605B	30.8±5.7735D	47.22±5.7735D	66±2.3094C	149.77±5.7735A	17.6
Total Nitrogen %	1.5±0.01732C	2.0267±0.02333B	2.36±0.02309A	2±0.02887B	2.06±0.01155B	0.055
Phosphorus %	0.2±0.00577A	0.2367±0.01202A	0.25±0.00577A	0.260.01155A	0.27±0.01155A	N. S
Potassium %	0.65±0.02887C	1.09±0.03464B	0.7±0.04041C	1.3±0.03464A	0.72±0.01155C	0.078
Sulfur mg/g.dry wt	10.59±1.15470D	22.5±1.1547C	22±1.1547C	27.67±1.1547B	31.27±1.1547A	2.95
Zinc mg/100g.f.wt	2.89±0.00577B	2.99±0.00577C	2.97±0.01155C	2.97±0.01732C	3±0.00577A	0.026
Iron%	1.32±0.01732C	1.78±0.02963B	1.17±0.01732D	2.35±0.02887A	1.77±0.01732B	0.066
Calcium mg/100g	29.05±0.05C	44.8±1.1547B	30.07±0.06506C	57.4±1.73205A	43.4±1.73205B	3.04
Fiber %	1.3333±0.15191A	1.27±0.05774A	1.66±0.06351A	1.85±0.0866A	1.6±0.02887A	N. S

From the observation of the results obtained in this study, conventional and nanofertilizers were more effective in increasing vegetative and physiological characteristics than the control treatment, in which only water was used, and there was also the superiority of nanofertilizers over conventional ones in most of the vegetative and physiological characteristics. These findings were consistent with the outcomes of [Salih and Kamall-Eldeen \(2024\)](#) who showed that the NPK spray treatment was superior in all the characteristics studied, giving the maximum number of flowering plants and the maximum nutrients values in the leaves. in terms of nitrogen, potassium and carbohydrate content. Also, consistent with the results of [Al-Tamimi et al. \(2023\)](#), who declared that foliar spraying of nanofertilizers increased the vegetative characteristics and chemical content of citrus plants, as the NPK nanofertilizers was superior by registering marked elevations in height, stem diameter, number of leaves, leaf area, shoot and root dry weight, root length, chlorophyll and carbohydrates, nitrogen and phosphorus.

This positive impact of nanofertilizers on the characteristics studied may be due to their dimensions of 30 to 40 nanometres and their ability to retain and slowly release ions because of their high surface area to the plant at the appropriate time [Rahale et al. \(2021\)](#). Thus, an increase in absorption rates and enhanced efficiency of photosynthesis can be achieved, resulting in an improvement in most vegetative characteristics of lettuce plants [Abdel-Hakim et al. \(2023\)](#).

Nanotechnology-based fertilizers are important because they are eco-friendly and contribute to sustainable agriculture. This technology makes it possible to use small particles of nanomaterials to create so-called smart fertilizer, which improves nutrient use efficiency and lowers the cost of environmental preservation [Bara'a and Abood \(2023\)](#). The impact of nanofertilizers in increasing plant growth and its chemical content is caused by the larger surface area and zones interaction on the surface of these nanofertilizers, as these characteristic facilitate the uptake of the fertilizers generated by the nanomaterials, or maybe due to their positive influence in meeting the plant's require for the main inorganic constituents required for, metabolic processes, respiration and photosynthesis, which increases the uptake of these elements by the plant when they are added. In addition, they play an important function in

cell division and evolution, as well as increasing root size, which contributes to efficient nutrient uptake [Nongbet et al. \(2022\)](#), and [Gade et al. \(2023\)](#).

Nitrogen is crucial for plant development and growth, particularly at the cell growth and division phase, so this increase in the vegetative and chemical content of the lettuce plant under study could be caused by the role of nitrogen as a component of amino acids, proteins, pigments, nucleic acids and enzymes [Leghari et al. \(2016\)](#), [Muhammad et al. \(2022\)](#). Also, Potassium has a direct role in activation of enzyme, maintenance of water status, translocation of assimilates, and protein synthesis and is involved in the regulation of cellular turgor pressure to prevent wilting, which in turn controls stomatal opening and thus increases drought tolerance [Hasanuzzaman et al. \(2018\)](#). Likewise, Phosphorus is a component of key molecules such as nucleic acids, phospholipids and ATPs. It is involved in the control of enzyme responses, photosynthesis, regulate metabolism, transfer energy, and the conversion of carbohydrates and protein formation leading to a developed in the vegetative growth of the plant [Tariq et al. \(2023\)](#).

Because of the importance of these three nutrients and their participation in many metabolic pathways within the plant, their presence in the nano-form increases their bioavailability within the plant's body and thus increases its growth and chemical content, and this is what was observed in the results of the current study. [Abdel-Aziz et al. \(2016\)](#) suggested that nano-engineered N, P and K fertilizers can improve the efficiency of nutrient absorbed and utilized by plants. Moreover, [Hong et al. \(2022\)](#) mentioned that lettuce plants fertilized with nitrogen, phosphorus and potassium were higher than those without fertilizer in growth, yield and quality from the height of the plant, the diameter of the stem, the yield and the content of vitamin C and dissolved sugars.

Conclusions and Recommendations

This study showed different effects of different fertilizer types and concentrations on lettuce growth and nutrient content. Nano NPK 4g/l treatment excelled in promoting leaf number, vitamin K, phosphorus, sulfur and zinc content. Conversely, a lower concentration of nano NPK (2g/l) maximized plant weight, leaf area and levels of carbohydrates, vitamins A and C, potassium, iron, calcium and fiber.

Whereas conventional NPK fertilizer outperformed all treatments in terms of plant height, total chlorophyll, carotenoid content, protein content and nitrogen content. These results suggest that the optimal fertilizer for lettuce cultivation depends on the specific growth parameter of interest.

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Novelty Statement

This research will provide valuable insights into the potential benefits of using nanofertilizers in hydroponic systems, leading to improved crop yield, and quality and reduced environmental impact.

Author's Contribution

Taiba F. Al-Mayyahi: Conceptualization and overall management, data collection and field research.

Dhafer A. Jameel: Wrote abstract, Introduction and methodology

Haider A. Alghanmi: Wrote results, discussion, conclusion and references.

Conflict of interest

The authors have declared no conflict of interest.

References

- Abdel-Aziz, H.M., M.N. Hasaneen and A.M. Omer. 2016. Nano chitosan-NPK fertilizer enhances the growth and productivity of wheat plants grown in sandy soil. *Spanish J. Agric. Res.*, 14: e0902-e0902. <https://doi.org/10.5424/sjar/2016141-8205>
- Abdel-Hakim, S.G., A.S. Shehata, S.A. Moghannem, M. Qadri, M.F.A. El-Ghany, E.A. Abdeldaym and O.S. Darwish. 2023. Nanoparticulate fertilizers increase nutrient absorption efficiency and agro-physiological properties of lettuce plant. *Agronomy*, 13: 691. <https://doi.org/10.3390/agronomy13030691>
- Al-Ani, M., L.U. Opara, D. Al-Bahri and N. Al-Rahbi. 2007. Spectrophotometric quantification of ascorbic acid contents of fruit and vegetables using the 2, 4-dinitrophenylhydrazine method. *J. Food Agric. Environ.*, 5: 165.
- Al-Kaisy, W.A. and B.A.H. Al-Mgadami. 2014. Effect of NPK fertilizer and root fertilizers Inicium on some physiological characters of *Lycopersicon esculentum* and its effect on mitotic division index of seedling radical apex. *Baghdad Sci. J.*, 11: 1441-1447. <https://doi.org/10.21123/bsj.2014.11.4.1441-1447>
- Al-Tamimi, H., S. Lateef and O. Mahmood. 2023. Effect of foliar spraying with Nano-NPK fertilizer in some growth characteristics and chemical content of some citrus rootstocks. *Rev. Bionatura*, 8: 116.
- Al-Zaidy, A. and R. Al-Ubaidy. 2017. Effect of adding wheat peat and spraying with its extract and organic nutrient vegeamino on growth and yield of red cabbage. *Iraqi J. Agric. Sci.*, 48: <https://doi.org/10.36103/ijas.v48i2.405>
- Bara'a, N. and M.R. Abood. 2023. Effect of nano and mineral NPK in vegetative growth parameter of pear saplings. *IOP conference series: Earth and environmental science*,
- Bender, M. and C. Wood. 2000. Total phosphorus in soil. *Methods of phosphorus analysis for soils, sediments, residuals, and waters*, 45:
- Bradford, M.M., 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal. Biochem.*, 72: 248-254. <https://doi.org/10.1006/abio.1976.9999>
- DuBois, M., K.A. Gilles, J.K. Hamilton, P.T. Rebers and F. Smith. 1956. Colorimetric method for determination of sugars and related substances. *Anal. Chem.*, 28: 350-356. <https://doi.org/10.1021/ac60111a017>
- Elemike, E.E., I.M. Uzoh, D.C. Onwudiwe and O.O. Babalola. 2019. The role of nanotechnology in the fortification of plant nutrients and improvement of crop production. *Appl. Sci.*, 9: 499. <https://doi.org/10.3390/app9030499>
- Fathi, A., 2022. Role of nitrogen (N) in plant growth, photosynthesis pigments, and N use efficiency: *A. Agrisost*, 28: 1-8.
- Gade, A., P. Ingle, U. Nimbalkar, M. Rai, R. Raut, M. Vedpathak, P. Jagtap and K.A. Abd-Elsalam. 2023. Nanofertilizers: The next generation of agrochemicals for long-term impact on sustainability in farming systems. *Agrochemicals*, 2: 257-278. <https://doi.org/10.3390/agrochemicals2020017>

- Hasanuzzaman, M., M.B. Bhuyan, K. Nahar, M.S. Hossain, J.A. Mahmud, M.S. Hossen, A.A.C. Masud, Moumita and M. Fujita. 2018. Potassium: A vital regulator of plant responses and tolerance to abiotic stresses. *Agronomy*, 8: 31. <https://doi.org/10.3390/agronomy8030031>
- Hassan, S.S., 1980. Spectrophotometric determination of the K vitamins. *Methods Enzymol. Elsevier.*, 67: 125-128. [https://doi.org/10.1016/S0076-6879\(80\)67018-9](https://doi.org/10.1016/S0076-6879(80)67018-9)
- Helaly, A.A., A.E. Ashmawi, A.A. Mohammed, M. El-Abd and A. Nofal. 2021. Effect of soil application of nano NPK fertilizers on growth, productivity and quality of Lettuce (*Lactuca sativa*). *Al-Azhar J. Agric. Res.*, 46: 91-100. <https://doi.org/10.21608/ajar.2021.218559>
- Hong, J., F. Xu, G. Chen, X. Huang, S. Wang, L. Du and G. Ding, 2022. Evaluation of the effects of nitrogen, phosphorus, and potassium applications on the growth, yield, and quality of lettuce (*Lactuca sativa* L.). *Agronomy*, 12: 2477. <https://doi.org/10.3390/agronomy12102477>
- Jadoon, S., A. Malik, M. Qazi and M. Aziz, 2013. Spectrophotometric method for the determination of vitamin A and E using Ferrozine-Fe (II) complex. *Asian J. Res. Chem.*, 6: 334-340.
- Jakhar, A.M., I. Aziz, A.R. Kaleri, M. Hasnain, G. Haider, J. Ma and Z. Abideen. 2022. Nano-fertilizers: A sustainable technology for improving crop nutrition and food security. *NanoImpact*, 27: 100411. <https://doi.org/10.1016/j.impact.2022.100411>
- Kirk, J. and R. Allen. 1965. Dependence of chloroplast pigment synthesis on protein synthesis: Effect of actidione. *Biochem. Biophys. Res. Commun.*, 21: 523-530. [https://doi.org/10.1016/0006-291X\(65\)90516-4](https://doi.org/10.1016/0006-291X(65)90516-4)
- Knoell, D.L., M.W. Julian, S. Bao, B. Besecker, J.E. Macre, G.D. Leikauf, R.A. DiSilvestro and E.D. Crouser. 2009. Zinc deficiency increases organ damage and mortality in a murine model of polymicrobial sepsis. *Crit. Care Med.*, 37: 1380-1388. <https://doi.org/10.1097/CCM.0b013e31819cfe4>
- Krüger, J., 2020. *Lactuca: Cultivation and uses.*
- Kumar, Y., T. Singh, R. Raliya and K. Tiwari. 2021. Nano fertilizers for sustainable crop production, higher nutrient use efficiency and enhanced profitability. *Indian J. Fert.*, 17: 1206-1214.
- Laghari, S.J., N.A. Wahocho, G.M. Laghari, A.H. Laghari, G.M. Bhabhan, K.H. Talpur, T.A. Bhutto, S.A. Wahocho and A.A. Lashari. 2016. Role of nitrogen for plant growth and development: A review. *Adv. Environ. Biol.*, 10: 209-219.
- Mackinney, G., 1941. Absorption of light by chlorophyll solutions. *J. Biol. Chem.*, 140: 315-322. [https://doi.org/10.1016/S0021-9258\(18\)51320-X](https://doi.org/10.1016/S0021-9258(18)51320-X)
- Mir, Y.H., S. Mir, M.A. Ganie, A.M. Shah, U. Majeed, M. Chesti, M. Mansoor, I. Irshad, A. Javed and S. Sadiq. 2022. Soilless farming: An innovative sustainable approach in agriculture. *Pharma Innov. J.*, 11: 2663-2675.
- Mostofa, M.G., M.M. Rahman, T.K. Ghosh, A.H. Kabir, M. Abdelrahman, M.A.R. Khan, K. Mochida and L.S.P. Tran. 2022. Potassium in plant physiological adaptation to abiotic stresses. *Plant Physiol. Biochem.*, 186: 279-289. <https://doi.org/10.1016/j.plaphy.2022.07.011>
- Mou, B., 2009. Nutrient content of lettuce and its improvement. *Curr. Nutr. Food Sci.*, 5: 242-248. <https://doi.org/10.2174/157340109790218030>
- Muhammad, I., L. Yang, S. Ahmad, S. Farooq, A.A. Al-Ghamdi, A. Khan, M. Zeeshan, M.S. Elshikh, A.M. Abbasi and X.B. Zhou. 2022. Nitrogen fertilizer modulates plant growth, chlorophyll pigments and enzymatic activities under different irrigation regimes. *Agronomy*, 12: 845. <https://doi.org/10.3390/agronomy12040845>
- Nicholls, J.W., J.P. Chin, T.A. Williams, T.M. Lenton, V. O'Flaherty and J.W. McGrath. 2023. On the potential roles of phosphorus in the early evolution of energy metabolism. *Front. Microbiol.*, 14: 1239189. <https://doi.org/10.3389/fmicb.2023.1239189>
- Nongbet, A., A.K. Mishra, Y.K. Mohanta, S. Mahanta, M.K. Ray, M. Khan, K.H. Baek and I. Chakrabartty. 2022. Nanofertilizers: A smart and sustainable attribute to modern agriculture. *Plants (Basel)*, 11: <https://doi.org/10.3390/plants11192587>
- Rahale, C.S., K. Subramanian and A. Lakshmanan. 2021. Nanofertilizer in enhancing the production potentials of crops. *Nanotechnology in plant growth promotion and protection: recent advances and impacts*, pp. 63-78. <https://doi.org/10.1002/9781119745884.ch4>
- Salih, Z.K. and A.A. Kamall-Eldeen. 2024. Manufacturing of NPK nano fertilizer and its

- effect on growth and of cut flowers production in two types of *Rosa* spp. *Exp. Theor. Nanotechnol.*, 8: 27–32. <https://doi.org/10.56053/8.2.27>
- Sandell, E.B., 1951. Colorimetric determinations of traces of metals. *LWW.* 71: 245. <https://doi.org/10.1097/00010694-195103000-00016>
- Saxena, M.M., 2001. Handbook of water and soil analysis. Nidhi Publishers (India).
- Shahane, A.A. and Y.S. Shivay. 2021. Soil health and its improvement through novel agronomic and innovative approaches. *Front. Agron.*, 3: 680456. <https://doi.org/10.3389/fagro.2021.680456>
- Shi, M., J. Gu, H. Wu, A. Rauf, T.B. Emran, Z. Khan, S. Mitra, A.S. Aljohani, F.A. Alhumaydhi and Y.S. Al-Awthan. 2022. Phytochemicals, nutrition, metabolism, bioavailability, and health benefits in lettuce. A comprehensive review. *Antioxidants*, 11: 1158. <https://doi.org/10.3390/antiox11061158>
- Singh, I. and N. Stoskopf. 1971. Harvest index in cereals 1. *Agron. J.*, 63: 224-226. <https://doi.org/10.2134/agronj1971.00021962006300020008x>
- Song, H., H. Wu and X. Li. 2018. Quantitative analysis of chemical components of plant fiber. *Appl. Chem. Eng.*,
- Sulaiman, J.Y. and S.M. Rasheed. 2024. Effect of foliar application by nano and non-nano NPK fertilizers on growth, yield and quality of two lettuce (*Lactuca sativa* L.) cultivars under plastic house conditions. *Acad. J. Nawroz Univ.*, 13: 105-116.
- Tariq, A., F. Zeng, C. Graciano, A. Ullah, S. Sadia, Z. Ahmed, G. Murtaza, K. Ismoilov and Z. Zhang. 2023. Regulation of metabolites by nutrients in plants. *Plant ionomics: Sensing, signaling, and regulation.* pp. 1-18. <https://doi.org/10.1002/9781119803041.ch1>
- Temminghoff, E.E. and V.J. Houba. 2004. Plant analysis procedures. Springer. <https://doi.org/10.1007/978-1-4020-2976-9>
- Tomar, N.S. and R. Agarwal. 2013. Influence of treatment of *Jatropha curcas* L. leachates and potassium on growth and phytochemical constituents of wheat (*Triticum aestivum* L.). <https://doi.org/10.4236/ajps.2013.45140>