Egypt. J. Soil Sci. **Vol. 64**, No. 3, pp: 927 – 950 (2024)

Egyptian Journal of Soil Science

http://ejss.journals.ekb.eg/

Enhancing Spinach Performance: Effectiveness of Nano-fertilizers in Conjunction with Conventional NPK Fertilizers

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THE WIDESPREAD application of conventional chemical fertilizers has led to various environmental issues, including reduced food quality, soil degradation, and harm to beneficial soil organisms. Conversely, the use of Nano-fertilizers holds promise as a potential solution to address these challenges. Therefore, the present study was conducted to evaluate the effectiveness of supplementing small quantities of Nano-fertilizers in conjunction with conventional NPK fertilizers to reduce the overall volume of traditional fertilizers when cultivating spinach plants. Moreover, the performance of micro- Nano fertilizers (Fe, Zn) with spinach plants was assessed in comparison to the performance of chelated fertilizers. A split-plot experimental design was utilized, consisting of twenty treatments with three replicates, which were the simple possible combination between four NPK additions as main plots and five foliar applications of Fe and Zn as sub main factor. Main factor treatments were **T_1:** 100% recommended dose of NPK as traditional bulk form, **T_2:** 75% recommended dose of NPK as traditional bulk form + 15% recommended dose of NPK as Nano form, **T_3:** 50% recommended dose of NPK as traditional bulk form + 15% recommended dose of NPK as Nano form and**T_4:** 25% recommended dose of NPK as Nano form. While, the sub main factor treatments were **F_1:** Control (without foliar)**, F_2:** Fe- Nano (10mg L^{-1}, Fe_2O_3 Nanoparticles)**, F_3:** Zn-Nano (10mg L^{-1}, ZnO Nanoparticles)**, F_4:** Fe- EDTA (100mg L^{-1} using Fe -EDTA 6% Fe) and **F_5:** Zn-EDTA (100mg L^{-1} using Zn -EDTA 6%Zn). The **T_2** treatment emerged as the most effective in achieving the highest values for all the all the measured parameters such as plant height (cm), fresh and dry weights (g $plant^{-1}$), leaf area (cm^2 $plant^{-1}$) and yield (ton ha^{-1}) of spinach plants at the harvest stage. It was followed closely by the **T_3** treatment. In contrast, the **T_1** treatment ranked third after both **T_3** and **T_2.** Lastly, the **T_4** treatment was found to have the least impact on these growth parameters and yield. Fe treatments exhibited greater effectiveness when compared to the Zn treatments. Furthermore, the data demonstrate that the Nano form exhibited greater effectiveness when compared to the chelated form in the context of both Fe and Zn treatments. Generally, it can be noticed that the **F_2** treatment was the superior for obtaining the maximum values for the most of studied traits. Therefore, it is recommended that farmers and agricultural practitioners consider adopting this approach to optimize crop performance while minimizing the environmental impact of conventional chemical fertilizers.

Keywords: Fe_2O_3 Nanoparticles, ZnO Nanoparticles, Fe –EDTA, Zn- EDTA.

1. Introduction

Continuous use of chemical fertilizers can lead to soil degradation. These fertilizers primarily provide three essential nutrients to plants however, they do not enhance soil structure. Over time, this can result in depleted soil fertility, decreased microbial activity, and reduced organic matter content (Elsherpiny and Faiyad, 2023). Excess application of chemical fertilizers can lead to nutrient runoff into nearby water bodies. Nitrogen and phosphorus, in particular, can cause eutrophication—a process where an excess of nutrients promotes algal growth. This can lead to oxygen depletion in water bodies, harming aquatic life and disrupting ecosystems. Additionally, nitrates from fertilizers can leach into groundwater, contaminating drinking water sources and posing health risks to humans and animals (Litskas 2023). Chemical fertilizers can contribute to air pollution

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Received: 21/03/2024; Accepted: 16/04/2024
DOI: 10.21608/EJSS.2024.278586.1741

through the release of ammonia (NH_3) and nitrous oxide (N_2O). Ammonia volatilization occurs when nitrogen fertilizers are applied to fields, leading to the release of ammonia gas into the atmosphere. Nitrous oxide is a potent greenhouse gas with a much higher warming potential than carbon dioxide. Its emission from fertilized soils contributes to climate change and stratospheric ozone depletion (Farouk *et al.* 2023). Intensive agricultural practices reliant on chemical fertilizers often involve the clearing of natural habitats, leading to habitat loss and fragmentation. Additionally, the disruption of soil ecosystems due to excessive fertilizer use can negatively impact soil-dwelling organisms, such as earthworms and beneficial microbes, leading to a decline in soil biodiversity (Kahandage *et al*. 2023). Generally, chemical fertilizers can leave residual effects on the environment, persisting in soil and water long after application. This can result in cumulative environmental damage over time, with consequences for both terrestrial and aquatic ecosystems (Elawady *et al*. 2024).

Nanotechnology has emerged as a transformative force across various domains of science and technology, offering groundbreaking solutions and innovations with the potential to reshape the way we approach challenges in fields ranging from medicine and electronics to agriculture and environmental conservation (Bhushan, 2017).

In this ever-evolving landscape, the utilization of nanotechnology holds particular promise in the realm of agriculture, where it plays a pivotal role in elevating the efficiency of fertilization. One of the key breakthroughs in this regard lies in the conversion of fertilizers, both for essential macro elements like nitrogen, phosphorus, and potassium (NPK), and vital microelements such as iron and zinc, into their Nano form (Preetha and Balakrishnan,2017; Hagab *et al.* 2018). This approach brings about a multitude of benefits that revolutionize the way we nourish our crops, enhancing nutrient delivery, optimizing plant nutrient uptake, and ultimately contributing to increased agricultural productivity, sustainable food production, and a more resilient environment (Rakhimol *et al.* 2021).

Nanoparticles have gained significant attention in the field of plant nutrition and agriculture due to their unique properties and potential benefits for enhancing plant growth and productivity (Elemike *et al.* 2019). Nanoparticles, especially metal and metal oxide nanoparticles, can be engineered to encapsulate or carry essential nutrients, such as micronutrients (*e.g.*, iron, zinc, and copper) and macronutrients (*e.g.*, nitrogen and phosphorus). These nanoparticles can protect the nutrients from leaching or chemical reactions in the soil and gradually release them to plants, promoting efficient nutrient uptake (Zhao *et al.* 2020).

Nanoparticles can improve nutrient uptake by plants due to their high surface area and reactivity. For example, zinc oxide nanoparticles have been shown to increase the bioavailability of zinc to plants, leading to better growth and increased crop yield (Mittal *et al.* 2020). Nanoparticles can also serve as carriers for pesticides and herbicides. Controlled release of agrochemicals through nanoparticles can reduce the overall amount of chemicals needed, minimize environmental contamination, and enhance their efficacy (Singh *et al.* 2021). Nanoparticles can be used to modify soil properties, enhancing its water-holding capacity, structure, and nutrient retention. This can lead to improved soil fertility and plant growth (Guleria *et al.* 2022).

It's important to note that the application of nanoparticles in agriculture also raises concerns about their potential toxicity to plants and the environment. Research is ongoing to understand the long-term effects of nanoparticles on soil, plant health, and surrounding ecosystems. The use of nanoparticles in agriculture is subject to regulatory oversight, and ethical considerations regarding their impact on the environment and human health are important aspects of research and application (Paramo *et al.* 2020).

Not long ago, the introduction of chelated fertilizers represented a significant advancement in the realm of foliar fertilization, as they offered the capability to transport nutrients to specific locations within the plant more rapidly than conventional mineral fertilizers. However, in the present era, the nanotechnology approach has risen to prominence as the superior method (Roosta *et al.* 2015). Nanotechnology provides a more precise and efficient mean of delivering nutrients to plants, ensuring they receive the right nutrients at the right time, ultimately outperforming the earlier chelated fertilization techniques (Mahdieh *et al.* 2018).

The superiority of the nanotechnology approach compared to the chelated approach lies in its precision and efficiency in delivering nutrients to plants. Nanotechnology allows for the controlled release of nutrients at the nanoscale, ensuring that plants receive the right amount of nutrients when they need them. This targeted delivery enhances nutrient uptake, promotes plant growth, and minimizes wastage (Dhaliwal *et al.* 2021). In contrast, chelated approaches, while effective to some extent, may not offer the same level of precision and efficiency in nutrient delivery (El-Desouky *et al.* 2021). Overall, the nanotechnology approach provides a promising solution for optimizing nutrient delivery to plants, thereby potentially improving crop yield and reducing environmental impacts compared to traditional chelated methods.

Nano-fertilizers offer potential advantages over traditional fertilizers due to their ability to enhance nutrient uptake by plants, reduce environmental impact through targeted delivery and decreased nutrient runoff, improve soil health by promoting microbial activity and nutrient stability, and integrate with other agricultural technologies for optimized nutrient management (Rakhimol *et al.* 2021). Their small size and tailored nutrient release mechanisms make them promising tools for increasing agricultural productivity while minimizing adverse effects on the environment (Singh *et al.* 2021).

Spinach (*Spinacia oleracea* L) stands as a nutritional powerhouse with immense health benefits. Rich in essential nutrients such as iron, vitamin C, and folate, it plays a crucial role in promoting overall well-being (Turan *et al.* 2022). Its versatility in culinary applications makes it a dietary staple, while its economic importance is significant for both local consumption and export markets, offering a source of income for farmers. Moreover, its nutritional content makes it a valuable addition to diets, supporting optimal health and addressing nutritional deficiencies, underscoring its vital role in both the food industry and public health (Sun *et al.* 2023).

Overall, while the application of nanoparticles in plant nutrition and agriculture holds promise for improving crop yields and sustainability, more research is needed to fully understand their effects and ensure responsible and safe use. Consequently, the present study is conducted with the objective of assessing how spinach plants respond to diverse mineral nutrient forms, including granular, Nano, and chelated, and identifying the most efficient form and application rate.

2. Material and Methods

Experimental site

A field research investigation was carried out at the Agriculture Faculty Farm of Mansoura University, Egypt, during the 2021/2022 and 2022/2023 growing seasons, located at coordinates 31°03′00″N 31°22′59″E.

Experimental design and treatments

A split-plot experimental design was utilized, consisting of twenty treatments with three replicates, which were the simple possible combination between four NPK additions as main plots and five foliar applications of Fe and Zn as sub main factor. The concentrations of nano-fertilizers and their application methods, as well as the selection of NPK ratios, were determined in accordance with the methodologies outlined in the studies conducted by El-Desouky *et al.* (2021) and Sharaf-Eldin *et al.* (2022). The NPK fertilizer underwent a transformation from its conventional composition (urea, phosphoric acid, and potassium sulfate) to a nanostructured form through the process of grinding. Utilizing a specialized nano-grinding mill, the standard NPK fertilizer was fragmented into finer particles, aiming to achieve a nanoscale size. Physical methodologies including ball milling were employed to further diminish particle dimensions, thereby facilitating the disruption of chemical bonds and augmenting material surface area. Throughout the grinding procedure, careful oversight of parameters such as rotation speed, grinding duration, and the ratio of grinding balls was maintained to ensure the attainment of uniformly sized nanoparticles with an even distribution. Additionally, optimal temperature and humidity levels were upheld during grinding to mitigate undesired outcomes such as moisture aggregation or unintended chemical reactions. The specific size requirements for nanoparticles were duly ascertained. Figure 1 displays the experiment's flowchart, representing the studied treatments using the following symbols:

Main factor

T_1: 100% recommended dose of NPK as traditional bulk form

T_2: 75% recommended dose of NPK as traditional bulk form + 15% recommended dose of NPK as Nano form

T_3: 50% recommended dose of NPK as traditional bulk form + 15% recommended dose of NPK as Nano form

T_4: 25% recommended dose of NPK as Nano form

Sub main factor

F_1: Control (without foliar)

F_2: Fe- Nano (10mg L^{-1}, Fe_2O_3 Nanoparticles)

F_3: Zn- Nano (10mg L^{-1}, ZnO Nanoparticles)

F_4: Fe- EDTA (100mg L^{-1} using Fe -EDTA 6% Fe)

F_5: Zn- EDTA (100mg L^{-1} using Zn -EDTA 6%Zn)

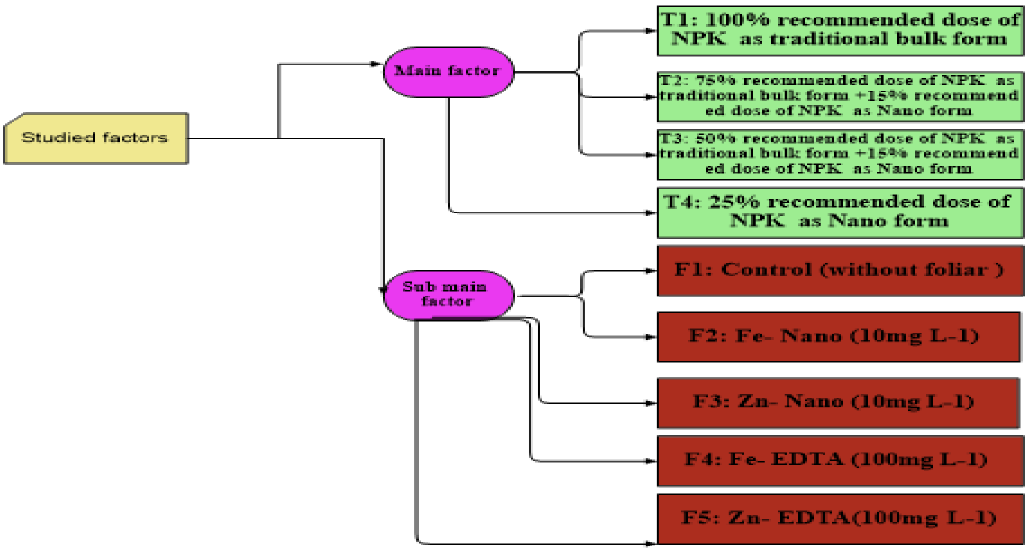


Fig. 1. Flowchart of the experiment.

Soil sampling and analysis

Table 1 shows some physical and chemical characteristics of the initial soil. Before the sowing process, experimental soil samples were collected from a depth of 0-25 cm for analysis as routine work using the standard methods. The organic matter content (O.M) was quantified using the Walkley and Black method using $K_2Cr_2O_7$, $FeSO_4$, $KMnO_4$, $H_2C_2O_4.2H_2O$ and diphenylamine indicator as described by Hesse (1971). Electrical conductivity (EC) was measured in a saturated soil paste extract which was obtained through the free capillary attraction method as mentioned by Jackson, (1967). Soil pH was gauged in a soil suspension (1:2.5) as mentioned by Richards (1954).The distribution of particle sizes was determined employing the pipette method according to the procedure described by Gee and Baudet (1986).The classification of the soil's texture class was achieved through the use of a soil texture triangle (Moreno-Maroto *et al.,* 2022). Soil-available nitrogen (NH_4+ NO_3^-) extraction was done through the Kjeldahl method. This method involves the use of potassium sulfate (K_2SO_4, 1%), devarda alloy, and sulphamic acid (H_3NSO_3, 2%) to ensure accurate results and to remove potential interference from nitrite ions (NO_2^-) (Dewis and Freitas, 1970). Soil-available phosphorus extraction was done through the Olsen method using spectrophotometer. In this method, the soil was mixed with a solution of sodium bicarbonate ($NaHCO_3$). The typical extraction solution used is 0.5 M $NaHCO_3$ at a specific pH (usually around 8.5). Also, active carbon purified, sulphomolybdic solution and stannous chloride ($SnCl_2$) were used in Olsen method (Dewis and Freitas, 1970). Soil-available potassium extraction was done by flame photometer method using ammonium acetate ($NH_4CH_3CO_2$) as solution extraction (Dewis and Freitas, 1970). The extraction of soil-available iron and zinc was conducted using Diethylene Triamine Penta Acetic Acid (DTPA) and measured through atomic absorption, as described by Dewis and Freitas (1970). Field capacity (FC) and Saturation (SP) and, expressed as percentages, were calculated using the following equation:

$$\% SP = 2\% FC = 4\% \text{ wilting point (WP)}$$

Table 1. Properties of the initial soil.

Characteristics	Values	Characteristics	Values
Chemical traits		**Availability of nutrients**	
O.M	1.30, g $100g^{-1}$	**Available -N**	45.3, mg kg^{-1}
EC	3.75, dS m^{-1}	**Available -P**	7.05, mg kg^{-1}
pH	7.89	**Available -K**	202.6, mg kg^{-1}
Particle size distribution		**Available -Fe**	1.55, mg kg^{-1}
Sand	24.5, %	**Available -Zn**	0.75, mg kg^{-1}
Silt	25.5, %	**Hydro physical properties**	
Clay	50.0, %	**FC**	35, %
Texture class	**Clayey**	**SP**	70

Characterization of nanoparticles

All Nano particulate fertilizers were acquired from Nano Fab Technology Company situated in Cairo, Egypt, and were utilized in accordance with the manufacturer's guidelines.

NPK Nanoparticles

Figure 2 illustrates the particles size of Nano- NPK fertilizers by electronic microscope (TEM). It is worth noting that the fertilizer that was converted from the traditional bulk form to Nano form NPK was (20:20:20 fertilizer), as the sources were urea (46%N), phosphoric acid (13% P), and potassium sulfate (containing 39.8% K).

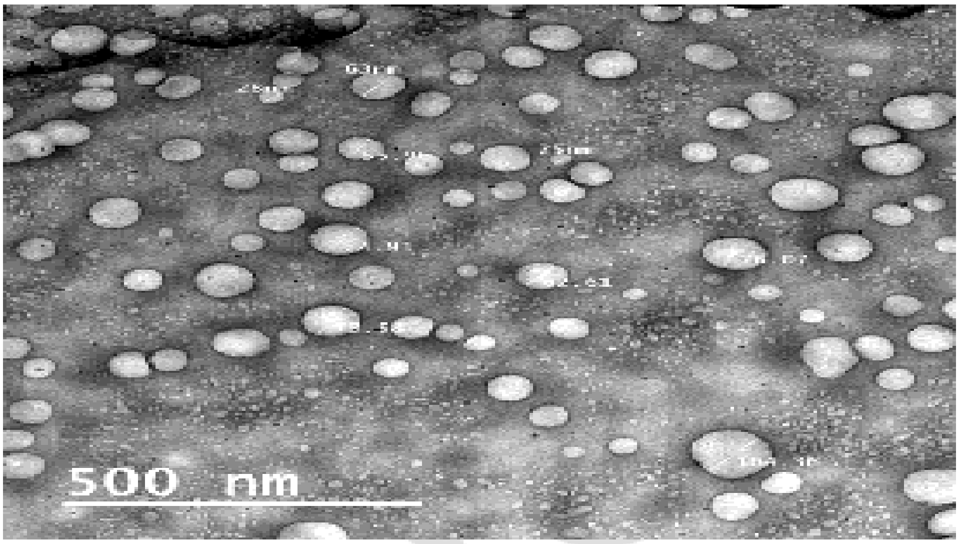


Fig. 2. Particles size of Nano- NPK fertilizers by electronic microscope.

Fe_2O_3 Nanoparticles

The physicochemical properties of Fe_2O_3 nanoparticles were assessed through TEM imaging (as shown in Figure 3). The images of the synthesized Fe_2O_3 nanoparticles exhibit a spherical morphology with an average particle size ranging from 12.5 to 17.5 nanometers.

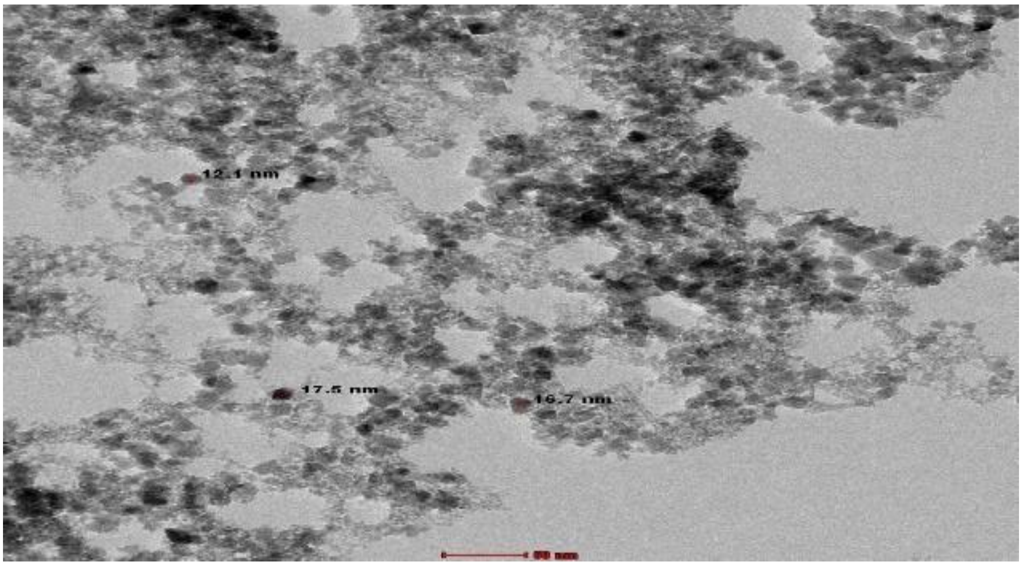


Fig. 3.TEM imaging of the prepared Fe_2O_3 nanoparticles.

ZnO Nanoparticles

The physicochemical properties of Zinc oxide nanoparticles were assessed through TEM imaging (as shown in Figure 4). The images of the synthesized ZnO nanoparticles exhibit a spherical morphology with an average particle size ranging from 12.9-29.0 nanometers.

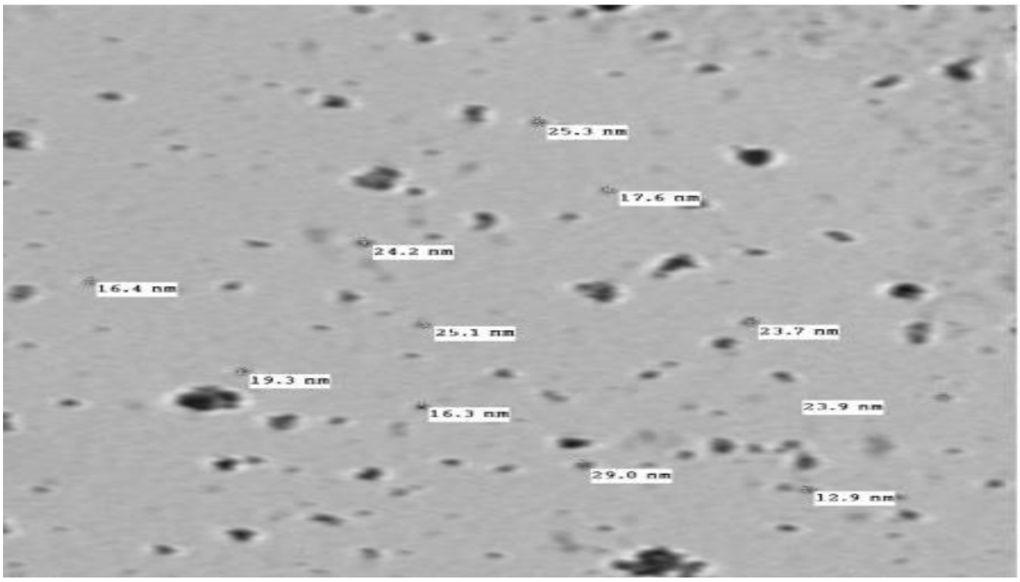


Fig. 4. TEM imaging of the prepared ZnO nanoparticles.

Spinach seeds

Seeds of the spinach variety "cv Nancy F1" were acquired from the Ministry of Agriculture and Soil Reclamation (MASR) and were planted during the first week of December in both of the studied seasons.

Cultivation and harvesting

According to the Ministry of Agriculture and Soil Reclamation (MASR) guidelines for spinach production, the recommended nutrient application rates are 40 units of nitrogen (N), 12 units of phosphorus (P), and 25 units of potassium (K) per feddan for NPK fertilizers. The recommended nutrient rates were applied in either traditional bulk form or Nano form, depending on the specific treatment under investigation. The traditional bulk form of N, P, and K was provided using ammonium sulfate (containing 21% N), calcium superphosphate (containing 6.7% P), and potassium sulfate (containing 39.8% K).

One month before cultivation, all plots (3.0m x 2.0m) were enriched with compost made from plant residues at a rate of 10 m^3 per feddan. Seeds were sown in rows at a rate of 6 kg per feddan. To accelerate germination and minimize the occurrence of soft wilt disease, the seeds were first soaked in water for 24 hours and allowed to dry. Subsequently, they were treated with a 0.75% thiram solution, and then promptly planted without any delay. Thinning of crowded plants was carried out so that the distance between plants became 5-10 cm, taking into account the elimination of weeds. For the traditional bulk form of NPK, calcium superphosphate was applied according to the specific treatments one month before cultivation. As for the traditional bulk forms of nitrogen (N) and potassium (K), they were administered based on the studied treatments in two equal doses. The first dose was provided three weeks after sowing, and the second dose was applied two weeks following the initial application.

In terms of NPK in Nano form, it was added according to the studied treatments two times in equal doses. The first dose was administered three weeks after sowing, and the second dose followed two weeks after the initial application.

Irrigation was conducted through surface irrigation methods, utilizing water sourced from the Nile River. The foliar application of iron (Fe) and zinc (Zn) in both studied forms was executed three times, with a 10-day interval between each application event. This series of applications commenced 30 days after sowing, and each application involved the use of 350 liters per feddan. Spinach was harvested at the pre-flowering stage, specifically when it reached the 5-6 leaf growth stage, approximately two months after sowing.

Measurement traits

Growth criteria and yield

During the harvest stage, which is also referred to as the marketing stage, representative samples of spinach plants were randomly selected. These samples were used to assess the vegetative growth performance, which was quantified in terms of plant height (cm), fresh and dry weights (g $plant^{-1}$), leaf area (cm^2 $plant^{-1}$) and yield (ton ha^{-1}).

Photosynthetic pigments

Also, at the same time, chlorophyll a &b and carotene (mg g^{-1}) were determined spectrometrically using acetone 80% as described by Porra *et al*. (1989).

Chemical composition and nutrient uptake

Also, at the harvest stage, the chemical composition of the entire spinach plant was determined. Nitrogen (N%), phosphorus (P%), potassium (K%), iron (Fe), and zinc (Zn) levels were evaluated using methods detailed by Walinga *et al.* (2013). Specifically, the Kjeldahl method was employed for N analysis, the spectrophotometric method for P, the flame photometer method for K, and the atomic absorption method for Fe and Zn. Prior to analysis, the samples underwent drying and digestion, using a 1:1 mixture of H_2SO_4 and $HClO_4$, following the procedure outlined by Peterburgski (1968). Nutrient uptake (including N, P, K, Fe, and Zn) in spinach leaves was quantified using the following formula:

Nutrient concentration (N, P, K, g $100g^{-1}$) in leaves **X** dry weight (g $plant^{-1}$)/100

Nutrient concentration (Fe, Zn, mg kg^{-1}) in leaves **X** dry weight (g $plant^{-1}$)/1000

Quality parameters

Vitamin C (mg $100g^{-1}$) was estimated spectrometrically using potassium permanganate solution, following the method outlined by Zanini *et al*. (2018). Nitrate (NO_3-N, mg kg^{-1}) was determined spectrometrically using a solution of 2% acetic acid and N-1naphthyle ethylene diamine dihydrochloride, following the procedure described by Singh, (1988). Oxalate (mg $100g^{-1}$) was assessed using the permanganometric method as detailed by Zhang *et al*. (2005). All quality traits were measured at the harvest stage.

Statistical analyses

Data were processed and analyzed using the COSTAT program (Version 6.303, CoHort, USA, 1998–2004). To identify significant differences between treatment means, the LSD (Least Significant Difference) test, as described by **Gomez and Gomez (1984),** was employed. Additionally, Duncan's Multiple Range test, as introduced by **Duncan (1955),** was utilized to enable the comparison of means among different treatments.

3. Results

Growth criteria and yield

Tables 2 and 3 present the influence of applying different NPK forms in various ratios (granular and Nano) and various Fe and Zn forms (Nano and chelated), as well as their interaction, on growth parameters such as plant height (cm), fresh and dry weights (g $plant^{-1}$) (Table 2), leaf area (cm^2 $plant^{-1}$) and yield (ton ha^{-1}) (Table 3) of spinach plants at the harvest stage.

Individual effect of NPK forms

Tables 2&3 highlight the significant impact of various NPK forms in different ratios on all the measured growth parameters, including plant height (cm), fresh and dry weights (g $plant^{-1}$), leaf area (cm^2 $plant^{-1}$) and yield (ton ha^{-1}) of spinach plants at the harvest stage.

Notably, the $\mathbf{T_2}$ treatment (75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form) emerged as the most effective in achieving the highest values for all the mentioned traits. It was followed closely by the $\mathbf{T_3}$ treatment (50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form). In contrast, the $\mathbf{T_1}$ treatment (100% recommended of NPK as traditional bulk form) ranked third after both $\mathbf{T_3}$ and $\mathbf{T_2}$**.** Lastly, the $\mathbf{T_4}$ treatment (25% recommended of NPK as Nano form) was found to have the least impact on these growth parameters and yield.

Individual effect of Fe and Zn forms

Except fresh weights (g $plant^{-1}$) which were non-significantly affected due to the studied foliar treatments, Tables 2&3 reveal that different forms of Fe and Zn (Nano and chelated) led to a significant improvement in all the measured parameters, including plant height (cm), dry weights (g $plant^{-1}$), leaf area (cm^2 $plant^{-1}$) and yield (ton ha^{-1}) of spinach plants at the harvest stage, as compared to the control group ($\mathbf{F_1}$) that did not receive any foliar application. This underscores the positive impact of using these Fe and Zn forms on spinach plant growth and productivity. Fe treatments exhibited greater effectiveness when compared to the Zn treatments. Furthermore, the data demonstrate that the Nano form exhibited greater effectiveness when compared to the chelated form in the context of both Fe and Zn treatments. Generally, it can be noticed that the $\mathbf{F_2}$ treatment **(**Fe- Nano at rate of 10mg L^{-1}) was the superior for obtaining the maximum values for all aforementioned traits followed by $\mathbf{F_3}$ treatment **(**Zn- Nano at rate of 10mg L^{-1}) then $\mathbf{F_4}$ treatment **(**Fe-EDTA at rate of 100mg L^{-1}) and $\mathbf{F_5}$ treatment **(**Zn-EDTA at rate of 100mg L^{-1}), respectively. While $\mathbf{F_1}$ treatment **(**without foliar application) came in the last order.

Table 2. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the growth parameters and yield of spinach plants (plant height, fresh and dry weights) at the harvest stage (combined data over both seasons).

Treatments		Plant height, cm	Fresh weight, g plant^{-1}	Dry weight, g plant^{-1}
Main factor: Form of NPK recommended dose				
T_1		30.72b	40.34c	8.01c
T_2		32.80a	43.21a	9.25a
T_3		32.32a	41.21b	9.01b
T_4		25.80c	34.36d	6.76d
LSD at 5%		**1.27**	**4.83**	**0.05**
Sub main factor: Foliar application treatments				
F_1		28.55b	37.57a	7.23e
F_2		31.31a	41.85a	8.70a
F_3		31.05a	37.98a	8.57b
F_4		30.69a	40.98a	8.45c
F_5		30.46a	40.51a	8.32d
LSD at 5%		**1.17**	**N.S***	**0.09**
Interaction				
	F_1	29.17	38.58	7.39
	F_2	31.83	41.85	8.41
T_1	**F_3**	31.27	41.10	8.25
	F_4	30.79	40.48	8.06
	F_5	30.53	39.68	7.92
	F_1	30.26	39.53	7.71
	F_2	33.55	45.34	9.72
T_2	**F_3**	33.51	31.43	9.65
	F_4	33.41	44.91	9.60
	F_5	33.28	44.82	9.55
	F_1	29.92	39.27	7.58
	F_2	33.08	44.45	9.48
T_3	**F_3**	33.06	44.23	9.39
	F_4	32.76	44.20	9.32
	F_5	32.78	43.89	9.25
	F_1	24.85	32.88	6.23
	F_2	26.77	35.76	7.18
T_4	**F_3**	26.35	35.17	7.00
	F_4	25.78	34.34	6.81
	F_5	25.23	33.66	6.55
LSD at 5%		**2.34**	**9.00**	**0.19**

Means within a column followed by a different letter (s) are statistically different at 5%

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**; T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1})

Table 3. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the growth parameters and yield of spinach plants (leaf area and yield) at the harvest stage (combined data over both seasons).

Treatments		Leaf area, cm^2 $plant^{-1}$	Yield, ton ha^{-1}
Main factor: Form of NPK recommended dose			
T_1		774.13c	6.32c
T_2		826.61a	6.85a
T_3		817.23b	6.69b
T_4		686.69d	4.81d
LSD at 5%		**4.66**	**0.08**
Sub main factor: Foliar application treatments			
F_1		735.41d	5.62e
F_2		795.26a	6.45a
F_3		789.47ab	6.35b
F_4		783.63bc	6.26c
F_5		777.05c	6.17d
LSD at 5%		**8.63**	**0.07**
Interaction			
	F_1	755.43	5.88
	F_2	792.20	6.61
T_1	**F_3**	782.70	6.50
	F_4	774.87	6.37
	F_5	765.43	6.24
	F_1	760.53	6.12
	F_2	847.30	7.10
T_2	**F_3**	845.90	7.04
	F_4	841.03	7.01
	F_5	838.27	7.00
	F_1	757.73	6.00
	F_2	835.20	6.95
T_3	**F_3**	833.57	6.88
	F_4	831.47	6.85
	F_5	828.20	6.78
	F_1	667.93	4.48
	F_2	706.33	5.14
T_4	**F_3**	695.70	4.98
	F_4	687.17	4.79
	F_5	676.30	4.66
LSD at 5%		**17.26**	**0.14**

Means within a column followed by a different letter (s) are statistically different at 5%

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**;** **T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1})

Interaction among the studied treatments

Regarding the interaction effect, the combined treatment of **T_2** treatment (75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form) x **F_2** treatment **(**Fe-Nano at rate of 10mg L^{-1}) realized the highest values of plant height (cm), fresh and dry weights (g $plant^{-1}$), leaf area (cm^2 $plant^{-1}$) and yield (ton ha^{-1}) of spinach plants at the harvest stage (Tables 2, 3), while the lowest values were noticed when spinach plants treated with 25% recommended of NPK as Nano form (**T_4** treatment) and simultaneously without foliar application (**F_1** treatment).

Chemical constituents

- Photosynthetic pigments

Table 4 displays the impact of employing various combinations of NPK forms (both in granular and Nano forms) and different forms of Fe and Zn (Nano and chelated), along with their interaction (Table 4), on the levels of photosynthetic pigments in spinach plants, specifically chlorophyll a and b as well as carotene (mg g^{-1}) at the time of harvest.

Individual effect of NPK forms

Table 4 shows that various NPK forms and ratios significantly affected the levels of photosynthetic pigments in spinach plants, specifically chlorophyll a and b as well as carotene (mg g^{-1}) at the harvest stage. Notably, the **T_2** treatment, which comprised 75% of the recommended NPK dose in traditional bulk form and 15% in Nano form, demonstrated the most effective outcome, resulting in the highest pigment values. Following closely was the **T_3** treatment, which consisted of 50% of the recommended NPK dose in traditional bulk form and 15% in Nano form.

In contrast, the **T_1** treatment, involving 100% of the recommended NPK in traditional bulk form, ranked third, trailing behind both **T_3** and **T_2.** Lastly, the **T_4** treatment, with only 25% of the recommended NPK in Nano form, exhibited the least impact on the levels of photosynthetic pigments in spinach plants.

Individual effect of Fe and Zn forms

Table 4 demonstrates a substantial enhancement in the photosynthetic pigment levels of spinach plants, including chlorophyll a and b, as well as carotene (mg g^{-1}) at the time of harvest, with the application of different forms of Fe and Zn, both Nano and chelated, in comparison to the control group (**F_1**), which lacked foliar treatment. This improvement is attributed to the supplementary Fe and Zn nutrients provided through Nano and chelated forms, which are more readily absorbed by plant tissues, thereby bolstering the plant's photosynthetic activity and pigment production, resulting in healthier and more vibrant spinach plants at the harvest stage.

Additionally, the data highlights that in both the Fe and Zn treatments, the Nano form outperformed the chelated form in terms of effectiveness, underscoring the superior impact of Nano-based nutrient delivery in promoting photosynthetic pigment levels in spinach plants.

Generally, a clear pattern emerges where the **F_2** treatment, involving Fe in Nano form at a rate of 10mg L^{-1}, demonstrated superior performance by recording the highest values for all the mentioned traits. Following closely was the **F_3** treatment, which utilized Zn in Nano form at a rate of 10mg L^{-1}. The **F_4** treatment, featuring Fe in the EDTA chelated form at a rate of 100mg L^{-1}, ranked third, while the **F_5** treatment, using Zn in the EDTA chelated form at the same rate, came in fourth place. Notably, the **F_1** treatment, which did not receive any foliar application, realized the lowest values for the assessed traits.

Interaction among the studied treatments

Concerning the interaction effect, the combined application of the **T_2** treatment (comprising 75% of the recommended NPK dose in traditional bulk form and 15% in Nano form) with the **F_2** treatment (involving Fe in Nano form at a rate of 10mg L^{-1}) resulted in the highest recorded values for chlorophyll a and b, as well as carotene (mg g^{-1}) in spinach plants at the time of harvest (as shown in Table 4). In contrast, the lowest values were observed when spinach plants were subjected to the **T_4** treatment, which provided 25% of the recommended NPK dose in Nano form, while simultaneously not receiving any foliar application (**F_1** treatment).

Table 4. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the photosynthetic pigments of spinach plants at the harvest stage (combined data over both seasons).

Treatments		Chlorophyll a	Chlorophyll b	Carotene
			(mg g^{-1})	
Main factor: Form of NPK recommended dose				
T_1		0.879c	0.710b	0.479c
T_2		0.922a	0.755a	0.508a
T_3		0.909b	0.742a	0.498b
T_4		0.789d	0.605c	0.436d
LSD at 5%		**0.007**	**0.014**	**0.003**
Sub main factor: Foliar application treatments				
F_1		0.830d	0.657e	0.454c
F_2		0.897a	0.725a	0.493a
F_3		0.889ab	0.719b	0.489a
F_4		0.882bc	0.711c	0.484b
F_5		0.876d	0.704d	0.481b
LSD at 5%		**0.009**	**0.005**	**0.004**
Interaction				
	F_1	0.840	0.673	0.458
	F_2	0.905	0.735	0.492
T_1	**F_3**	0.891	0.725	0.487
	F_4	0.885	0.712	0.481
	F_5	0.874	0.704	0.477
	F_1	0.861	0.694	0.471
	F_2	0.942	0.775	0.521
T_2	**F_3**	0.939	0.772	0.519
	F_4	0.936	0.770	0.515
	F_5	0.932	0.765	0.512
	F_1	0.851	0.681	0.466
	F_2	0.930	0.761	0.509
T_3	**F_3**	0.925	0.760	0.508
	F_4	0.921	0.757	0.505
	F_5	0.919	0.752	0.503
	F_1	0.768	0.580	0.420
	F_2	0.811	0.628	0.449
T_4	**F_3**	0.801	0.618	0.443
	F_4	0.788	0.605	0.437
	F_5	0.777	0.594	0.433
LSD at 5%		**0.018**	**0.010**	**0.009**

Means within a column followed by a different letter (s) are statistically different at 5%

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**;** **T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1})

Nutritional element concentration and uptake

Data of Tables 5, 6, 7 and 8 point out the individual impact of the NPK treatments and foliar application treatments (both Fe and Zn) as well as their interaction effect on nutrient concentrations in leaves of spinach plants *i.e.,* N, P, K (%) Fe, Zn (mgkg^{-1}) and nutrient uptake *i.e.,* N, P, K (g plant^{-1}), Fe, Zn (mg plant^{-1}).

Individual effect of NPK forms

Data presented in the same Tables indicate that the NPK treatments significantly affected the values of nutrient concentrations in leaves of spinach plants *i.e.,* N, P, K (%) (Table 5) Fe, Zn (mg kg^{-1}) (Table 6) and nutrient uptake *i.e.,* N, P, K (g plant^{-1}) (Table7) Fe, Zn (mg plant^{-1}) (Table 8) at the harvest stage. In can be noticed that the sequence order from the most effective treatment to less was as follows;

$\mathbf{T_2}$ treatment, comprising 75% of the recommended NPK dose in traditional bulk form and 15% in Nano form followed by the $\mathbf{T_3}$ treatment, which involved 50% of the recommended NPK dose in traditional bulk form and 15% in Nano form then $\mathbf{T_1}$ treatment, utilizing 100% of the recommended NPK in traditional bulk form and lately the $\mathbf{T_4}$ treatment, incorporating only 25% of the recommended NPK in Nano form.

Individual effect of Fe and Zn forms

Also, the same Tables demonstrate a substantial enhancement in the values of nutrient concentrations in leaves of spinach plants *i.e.,* N, P, K (%) (Table 5) Fe, Zn (mg kg^{-1}) (Table 6) and nutrient uptake *i.e.,* N, P, K (g plant^{-1}) (Table7) Fe, Zn (mg plant^{-1}) (Table 8) at the harvest stage. This enhancement is associated with the application of various forms of Fe and Zn, including Nano and chelated forms, when compared to the control group **($\mathbf{F_1}$)** that did not receive any foliar treatment. Generally, it can be noticed that the superior treatment for obtaining the maximum values of the studied nutrient concentrations and their uptake was $\mathbf{F_2}$ treatment, involving Fe in Nano form at a rate of 10mg L^{-1}, followed by the $\mathbf{F_3}$ treatment, which utilized Zn in Nano form at a rate of 10mg L^{-1}, then the $\mathbf{F_4}$ treatment, featuring Fe in the EDTA chelated form at a rate of 100mg L^{-1},then the $\mathbf{F_5}$ treatment, using Zn in the EDTA chelated form at the same rate and lately the $\mathbf{F_1}$ treatment, which did not receive any foliar application.

Interaction among the studied treatments

In terms of the interaction effect, the combined application of the $\mathbf{T_2}$ treatment (consisting of 75% of the recommended NPK dose in traditional bulk form and 15% in Nano form) with the $\mathbf{F_2}$ treatment (involving Fe in Nano form at a rate of 10mg L^{-1}) resulted in the highest recorded values for nutrient concentrations in leaves of spinach plants *i.e.,* N, P, K (%) (Table 5) Fe, Zn (mg kg^{-1}) (Table 6) and nutrient uptake *i.e.,* N, P, K (g plant^{-1}) (Table7) Fe, Zn (mg plant^{-1}) (Table 8) at the harvest stage. In contrast, the lowest values were observed when spinach plants were subjected to the $\mathbf{T_4}$ treatment, which provided only 25% of the recommended NPK dose in Nano form, while simultaneously not receiving any foliar application ($\mathbf{F_1}$ treatment).

Table 5. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the nutritional element concentration in leaves of spinach plants (N, P, K) at the harvest stage (combined data over both seasons).

Treatments		Nitrogen (N)	Phosphorus (P)	Potassium (K)
			(%)	
Main factor: Form of NPK recommended dose				
T_1		3.25c	0.456c	2.95c
T_2		3.50a	0.480a	3.16a
T_3		3.41b	0.474b	3.08b
T_4		2.83d	0.410d	2.52d
LSD at 5%		**0.08**	**0.004**	**0.06**
Sub main factor: Foliar application treatments				
F_1		3.02c	0.432d	2.71c
F_2		3.36a	0.466a	3.04a
F_3		3.32ab	0.463ab	3.00ab
F_4		3.29b	0.459bc	2.96ab
F_5		3.25b	0.456c	2.93b
LSD at 5%		**0.07**	**0.005**	**0.10**
Interaction				
T_1	**F_1**	3.07	0.438	2.74
	F_2	3.38	0.469	3.09
	F_3	3.31	0.464	3.03
	F_4	3.28	0.458	2.98
	F_5	3.22	0.454	2.93
T_2	**F_1**	3.16	0.448	2.88
	F_2	3.62	0.491	3.27
	F_3	3.60	0.489	3.25
	F_4	3.57	0.488	3.21
	F_5	3.55	0.485	3.19
T_3	**F_1**	3.12	0.443	2.78
	F_2	3.51	0.485	3.18
	F_3	3.49	0.483	3.15
	F_4	3.48	0.480	3.16
	F_5	3.46	0.479	3.12
T_4	**F_1**	2.73	0.397	2.43
	F_2	2.95	0.420	2.61
	F_3	2.87	0.416	2.57
	F_4	2.83	0.410	2.50
	F_5	2.77	0.405	2.48
LSD at 5%		**0.15**	**0.009**	**0.21**

Means within a column followed by a different letter (s) are statistically different at 5%

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**; T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1}).

Table 6. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the nutritional element concentration in leaves of spinach plants (Fe, Zn) at the harvest stage (combined data over both seasons).

Treatments		Iron (Fe)	Zinc (Zn)
		(mgkg^{-1})	
Main factor: Form of NPK recommended dose			
T_1		77.31c	50.61c
T_2		79.11a	52.11a
T_3		78.21b	51.37b
T_4		75.65d	49.28d
LSD at 5%		**0.48**	**0.57**
Sub main factor: Foliar application treatments			
F_1		74.38c	48.14d
F_2		80.79a	49.74c
F_3		76.43b	53.77a
F_4		80.36a	49.31c
F_5		75.89b	53.27b
LSD at 5%		**0.96**	**0.50**
Interaction			
	F_1	74.42	48.23
	F_2	80.31	49.53
T_1	**F_3**	76.19	53.29
	F_4	79.93	49.16
	F_5	75.72	52.83
	F_1	75.45	48.73
	F_2	82.16	51.23
T_2	**F_3**	78.45	55.21
	F_4	81.68	50.70
	F_5	77.79	54.69
	F_1	74.87	48.47
	F_2	81.22	50.33
T_3	**F_3**	77.30	54.31
	F_4	80.88	49.94
	F_5	76.76	53.80
	F_1	72.78	47.10
	F_2	79.47	47.86
T_4	**F_3**	73.77	52.26
	F_4	78.95	47.44
	F_5	73.29	51.74
LSD at 5%		**1.71**	**0.99**

Means within a column followed by a different letter (s) are statistically different at 5%

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**;** **T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1}).

Table 7. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the nutritional element uptake by spinach plants (N, P, K) at the harvest stage (combined data over both seasons).

Treatments		Nitrogen (N)	Phosphorus (P)	Potassium (K)
			(g plant^{-1})	
Main factor: Form of NPK recommended dose				
T_1		0.261c	0.037c	0.237c
T_2		0.325a	0.045a	0.293a
T_3		0.308b	0.043b	0.278b
T_4		0.191d	0.028d	0.170d
LSD at 5%		**0.008**	**0.001**	**0.004**
Sub main factor: Foliar application treatments				
F_1		0.219d	0.031d	0.197d
F_2		0.295a	0.041a	0.267a
F_3		0.287b	0.040b	0.260ab
F_4		0.281bc	0.039b	0.253bc
F_5		0.274c	0.038c	0.247c
LSD at 5%		**0.007**	**0.001**	**0.009**
Interaction				
T_1	F_1	0.227	0.032	0.202
	F_2	0.284	0.039	0.260
	F_3	0.273	0.038	0.250
	F_4	0.264	0.037	0.240
	F_5	0.255	0.036	0.232
T_2	F_1	0.244	0.035	0.222
	F_2	0.352	0.048	0.318
	F_3	0.347	0.047	0.314
	F_4	0.343	0.047	0.308
	F_5	0.339	0.046	0.304
T_3	F_1	0.237	0.034	0.211
	F_2	0.333	0.046	0.301
	F_3	0.328	0.045	0.296
	F_4	0.324	0.045	0.295
	F_5	0.321	0.044	0.289
T_4	F_1	0.170	0.025	0.151
	F_2	0.212	0.030	0.188
	F_3	0.201	0.029	0.180
	F_4	0.193	0.028	0.170
	F_5	0.181	0.027	0.162
LSD at 5%		**0.014**	**0.001**	**0.018**

Means within a column followed by a different letter (s) are statistically different at 5%

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**; T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1}).

Table 8. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the nutritional element uptake by spinach plants (Fe, Zn) at the harvest stage (combined data over both seasons).

Treatments		Iron (Fe)	Zinc (Zn)
		(mg plant^{-1})	
Main factor: Form of NPK recommended dose			
T_1		0.620c	0.406c
T_2		0.733a	0.483a
T_3		0.706b	0.464b
T_4		0.512d	0.333d
LSD at 5%		**0.004**	**0.006**
Sub main factor: Foliar application treatments			
F_1		0.538e	0.348e
F_2		0.704a	0.434c
F_3		0.657c	0.462a
F_4		0.680b	0.418d
F_5		0.633d	0.444b
LSD at 5%		**0.011**	**0.006**
Interaction			
	F_1	0.550	0.356
	F_2	0.676	0.417
T_1	**F_3**	0.629	0.440
	F_4	0.645	0.396
	F_5	0.600	0.418
	F_1	0.582	0.376
	F_2	0.799	0.498
T_2	**F_3**	0.757	0.533
	F_4	0.784	0.487
	F_5	0.743	0.523
	F_1	0.568	0.368
	F_2	0.770	0.477
T_3	**F_3**	0.726	0.510
	F_4	0.754	0.465
	F_5	0.710	0.498
	F_1	0.453	0.293
	F_2	0.571	0.344l
T_4	**F_3**	0.517	0.366
	F_4	0.5381	0.323
	F_5	0.480	0.339
LSD at 5%		**0.024**	**0.013**

Means within a column followed by a different letter (s) are statistically different at 5%

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**;** **T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1})

Quality parameters

Table 9 shows the outcomes resulting from the utilization of different combinations of NPK (in both granular and Nano forms) and various forms of Fe and Zn (Nano and chelated), along with their interactions (Table 9 & Fig 1). The impact of these combinations is assessed concerning quality parameters, including vitamin C (mg 100g^{-1}), nitrate (NO_3-N, mg kg^{-1}) and oxalate (mg 100g^{-1}) at harvest stage.

Individual effect of NPK forms

In terms of vitamin C (mg 100g^{-1}), the Table 9 shows that the **T_2** treatment, which comprised 75% of the recommended NPK dose in traditional bulk form and 15% in Nano form, demonstrated the most effective outcome, resulting in the highest values. Following closely was the **T_3** treatment, which consisted of 50% of the recommended NPK dose in traditional bulk form and 15% in Nano form. In contrast, the **T_1** treatment, involving 100% of the recommended NPK in traditional bulk form, ranked third, trailing behind both **T_3** and **T_2.** Lastly, the **T_4** treatment, with only 25% of the recommended NPK in Nano form, exhibited the least impact on the levels of vitamin C in spinach plants.

Table 9. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the quality parameters of spinach plants at the harvest stage (combined data over both seasons).

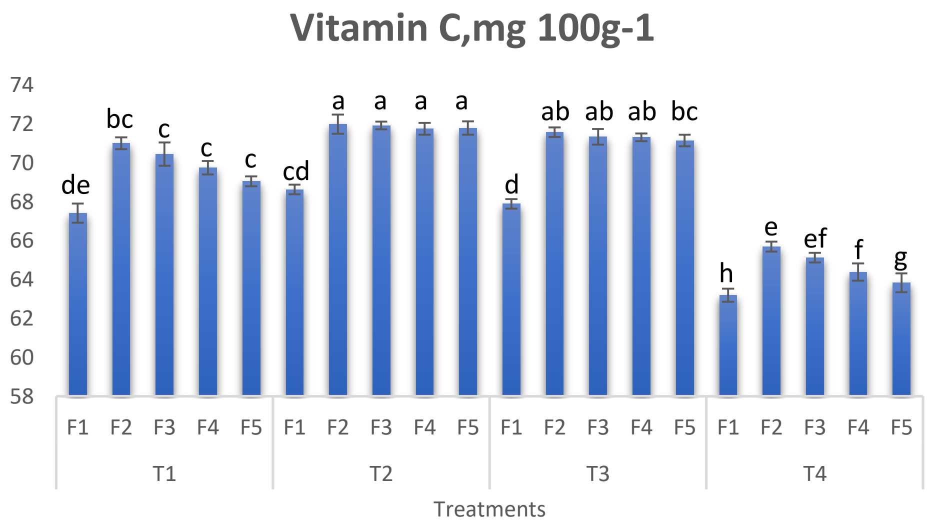
Treatments		Vitamin C, mg $100g^{-1}$	NO_3, mg kg^{-1}	Oxalate, mg $100g^{-1}$
Main factor: Form of NPK recommended dose				
T_1		69.55b	441.15a	792.73a
T_2		71.23a	381.39b	713.09b
T_3		70.66a	323.42c	633.86c
T_4		64.45c	263.35d	554.67d
LSD at 5%		**0.97**	**3.07**	**6.57**
Sub main factor: Foliar application treatments				
F_1		66.79d	328.75e	642.05e
F_2		70.08a	375.73a	705.26a
F_3		69.72ab	364.40b	689.00b
F_4		69.31bc	352.35c	673.88c
F_5		68.97c	340.41d	657.74d
LSD at 5%		**0.74**	**3.71**	**7.47**
Interaction				
	F_1	67.42	417.27	761.97
	F_2	71.02	465.50	824.17
T_1	**F_3**	70.46	453.43	808.00
	F_4	69.76	440.80	793.47
	F_5	69.06	428.73	776.03
	F_1	68.64	358.47	680.93
	F_2	72.00	404.23	744.97
T_2	**F_3**	71.93	392.67	728.87
	F_4	71.76	382.17	713.53
	F_5	71.80	369.40	697.13
	F_1	67.90	299.80	602.67
	F_2	71.59	346.27	665.27
T_3	**F_3**	71.35	335.87	649.13
	F_4	71.32	323.23	633.77
	F_5	71.16	311.93	618.47
	F_1	63.20	239.47	522.63
	F_2	65.70	286.90	586.63
T_4	**F_3**	65.13	275.63	570.00
	F_4	64.39	263.20	554.73
	F_5	63.84	251.57	539.33
LSD at 5%		**1.49**	**7.41**	**14.96**

Means within a column followed by a different letter (s) are statistically different at 5%

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**;** **T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1}).

T_2 Treatment (75% NPK in Traditional Bulk Form and 15% in Nano Form) achieved the highest vitamin C content due to the optimized combination of traditional bulk NPK and Nano NPK, enhancing nutrient uptake and assimilation, resulting in improved vitamin C production. **T_3** treatment (50% NPK in Traditional Bulk Form and 15% in Nano Form): This treatment closely followed **T_2** because it also included a substantial portion of Nano NPK, which contributed to improved nutrient efficiency and higher vitamin C levels. **T_1** Treatment (100% NPK in Traditional Bulk Form) ranked lower as it relied solely on traditional bulk NPK, which might not have been as efficiently absorbed by the plants, leading to comparatively lower vitamin C content. **T_4** Treatment (25% NPK in Nano Form),with a minimal amount of Nano NPK, exhibited the least impact on vitamin C levels, possibly because Nano NPK alone was insufficient to support optimal plant nutrition for vitamin C production.

In terms of nitrate (NO_3-N, mg kg^{-1}) and oxalate (mg $100g^{-1}$), it can be noticed that their values increased as the NPK ratio in traditional bulk form increased. The highest nitrate and oxalate values were associated with the T_1 treatment, which contained 100% traditional bulk NPK. The nitrate (NO_3-N) concentration in the spinach varied from 251.57 to 465.50 mg kg^{-1} of fresh weight. These levels were below the acceptable NO_3-N content for human consumption, which typically ranges from 3500 to 4500 mg kg^{-1} of fresh weight, as indicated by Abdel-Hakim *et al.* (2023). This outcome suggests that traditional bulk NPK may contribute to higher nitrate and oxalate accumulation in the plant tissues, possibly due to the more abundant presence of these nutrients in the form that is easily taken up by plants.



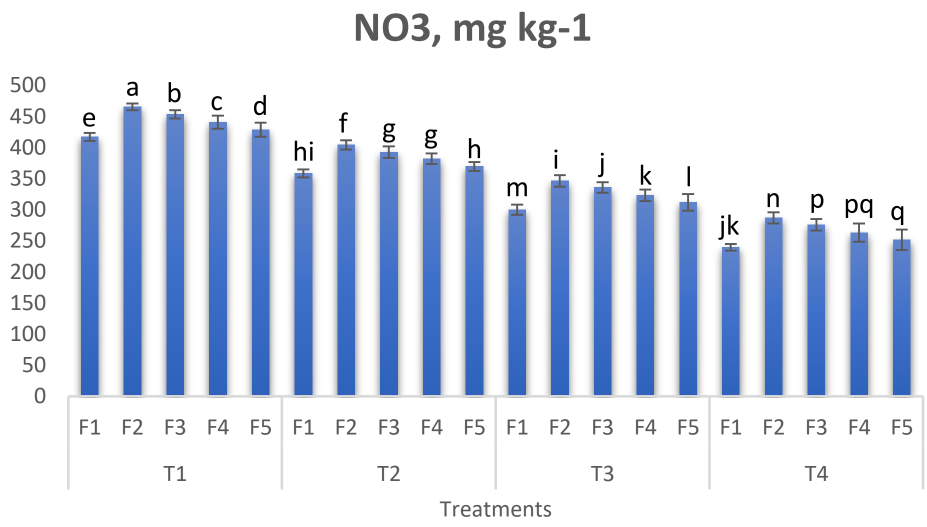


Fig. 6. The impact of various mineral nutrient forms (granular, Nano, or chelated) on the quality parameters of spinach plants at the harvest stage (combined data over both seasons).

Since, T_1: 100% recommended dose of NPK as traditional bulk form ; **T_2:** 75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form; **T_3:** 50% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form**;** **T_4:** 25% recommended dose of NPK as Nano form; **F_1:** Control (without foliar); **F_2:** Fe- Nano (10mg L^{-1}) ; **F_3:** Zn- Nano (10mg L^{-1}) **F_4:** Fe- EDTA (100mg L^{-1}); **F_5:** Zn- EDTA(100mg L^{-1}).

Individual effect of Fe and Zn forms

Table 9 reveals that different forms of Fe and Zn (Nano and chelated) led to a significant increase in all the measured parameters, including vitamin C (mg $100g^{-1}$), nitrate (NO_3-N, mg kg^{-1}) and oxalate (mg $100g^{-1}$) at harvest stage, as compared to the control group (F_1) that did not receive any foliar application. Also, the data demonstrate that the Nano form led to the highest values for vitamin C (mg $100g^{-1}$), nitrate (NO_3-N, mg kg^{-1}) and oxalate (mg $100g^{-1}$) when compared to the chelated form in the context of both Fe and Zn treatments. Generally, it can be noticed that the $\mathbf{F_2}$ treatment **(**Fe- Nano at rate of 10mg L^{-1}) was the superior for obtaining the maximum values for all aforementioned traits followed by $\mathbf{F_3}$ treatment **(** Zn- Nano at rate of 10mg L^{-1}) then $\mathbf{F_4}$ treatment **(**Fe- EDTA at rate of 100mg L^{-1}) and $\mathbf{F_5}$ treatment **(**Zn- EDTA at rate of 100mg L^{-1}), respectively. While $\mathbf{F_1}$ treatment **(**without foliar application) came in the last order**.**

Interaction among the studied treatments

In terms of vitamin C (mg $100g^{-1}$), the data in the same Table and Fig 2 show the combined treatment of $\mathbf{T_2}$ treatment (75% recommended dose of NPK as traditional bulk form +15% recommended dose of NPK as Nano form) x $\mathbf{F_2}$ treatment **(**Fe- Nano at rate of 10mg L^{-1}) realized the highest values, while the lowest values were noticed when spinach plants treated with 25% recommended of NPK as Nano form ($\mathbf{T_4}$ treatment) and simultaneously without foliar application ($\mathbf{F_1}$ treatment). The combination of a significant portion of NPK in traditional bulk form along with Nano NPK ($\mathbf{T_2}$) likely led to better nutrient availability and uptake, promoting higher vitamin C synthesis. The addition of Fe-Nano ($\mathbf{F_2}$) further enhanced nutrient absorption and utilization, resulting in the highest vitamin C content. Nano forms of nutrients often offer increased surface area and improved solubility, facilitating nutrient uptake by plant roots and subsequent utilization for metabolite production.

In terms of nitrate (NO_3-N, mg kg^{-1}) and oxalate (mg $100g^{-1}$), it can be noticed from the same Table that the combined treatment of $\mathbf{T_1}$ treatment (100% recommended dose of NPK as traditional bulk form) x $\mathbf{F_2}$ treatment **(**Fe- Nano at rate of 10mg L^{-1}) realized the highest values, while the lowest values were noticed when spinach plants treated with 25% recommended of NPK as Nano form ($\mathbf{T_4}$ treatment) and simultaneously without foliar application ($\mathbf{F_1}$ treatment). The highest nitrate and oxalate values were observed in this combination. When NPK is primarily in traditional bulk form ($\mathbf{T_1}$), it likely provides a substantial reservoir of nitrate and oxalate precursors that can be readily absorbed by plants, leading to higher accumulations of these compounds. The addition of Fe-Nano ($\mathbf{F_2}$) might have facilitated the uptake of nutrients and their conversion into nitrate and oxalate.

Generally, in spinach, nitrate and oxalate accumulation is particularly significant. As mineral nitrogen levels, especially in the form of nitrates, increase in the soil, spinach plants tend to absorb and accumulate these compounds. Nitrate is vital for spinach growth as it's used in the production of amino acids and proteins. However, excessive nitrate accumulation in spinach can lead to health concerns, such as potential nitrate toxicity for consumers. Additionally, spinach is known to accumulate oxalate, which can also increase with higher mineral nitrogen availability. The accumulation of oxalate in spinach can affect its nutritional quality and palatability, potentially leading to the perception of astringency in the leaves.

4. Discussion

The observed results can be attributed to several scientific reasons and insights. The $\mathbf{T_2}$ treatment, which included a combination of mineral and Nano forms, showed superior results. This could be due to the enhanced efficiency of nutrient uptake and utilization in this treatment. Nano-formulated nutrients have a higher surface area, which can lead to better absorption by plant roots, resulting in improved growth. The $\mathbf{T_3}$ treatment also performed well. This suggests that a balanced combination of nutrient sources can be effective. The synergy between mineral and Nano forms might provide a consistent supply of nutrients to the plants throughout their growth stages. The $\mathbf{T_1}$ treatment, which used solely mineral forms, ranked third. This result may be attributed to the slower release and lower efficiency of nutrient uptake associated with granular mineral forms. Plants may have experienced delayed access to nutrients, impacting their overall growth and yield. The $\mathbf{T_4}$ treatment, utilizing only 25% of the NPK dose as Nano form, exhibited the least favorable outcomes. This finding underscores that an insufficient amount of Nano-formulated nutrients may not provide the necessary nutrients for optimal plant growth (Rop *et al.* 2019).

In summary, the superior performance of the T_2 treatment may be attributed to the combination of mineral and Nano forms, which enhances nutrient uptake and utilization. The T_3 treatment also yielded positive results, demonstrating the advantages of balanced nutrient sourcing. These findings underscore the importance of nutrient source ratios and the potential benefits of incorporating Nano-formulated nutrients in optimizing the growth and yield of spinach plants. In general, the enhanced effectiveness of Nano fertilizers may be attributed to their diminutive particle size and expansive surface area, which amplify the capacity for nutrient absorption and uptake by plant roots. This, in turn, leads to more efficient nutrient utilization, consequently reducing the overall quantity of required fertilizers (Mali *et al.* 2020). Furthermore, the heightened mobility of nanoparticles within both soil and plant tissues facilitates the conveyance of nutrients throughout the plant, thereby enhancing nutrient dispersal and utilization. Finally, it can be said that the synergistic combination of traditional bulk or granular NPK fertilizers with Nano NPK was pivotal in boosting the growth and yield of spinach. Traditional fertilizers offer a reliable and steady nutrient supply, while Nano NPK, with its enhanced solubility and nutrient uptake, ensures efficient utilization of these nutrients. This balanced approach not only provides a consistent source of essential macronutrients but also allows for controlled, gradual nutrient release. Additionally, Nano NPK's ability to precisely target plant tissues optimizes nutrient utilization, leading to increased chlorophyll production and photosynthetic activity, ultimately resulting in higher spinach yields. The obtained results are in harmony with those of Gil-Díaz *et al.* (2022); Ibrahim, (2022).

Spinach, like many other plants, can suffer from Fe and Zn deficiencies, which can lead to stunted growth, reduced yield, and chlorosis (yellowing of leaves) (Barker and Pilbeam 2015). The foliar application of Fe and Zn forms addresses these deficiencies, leading to healthier and more productive plants. The positive impact of using Fe and Zn forms on spinach plant growth and productivity can be attributed to their essential roles in plant nutrition. While both elements are essential, the differential requirements of Fe and Zn in plant physiology, influenced by the specific experimental conditions and plant responses, might explain the observed greater effectiveness of Fe treatments compared to Zn treatments in this particular study.

The application of Fe and Zn treatments as foliar sprays on the leaves enhances their direct accessibility to the plant's foliage. This method allows for the rapid absorption of nutrients through the leaf surface, which is rich in stomata and cuticles, facilitating efficient nutrient uptake. The Nano form's smaller particle size, in this context, plays a crucial role in increasing the effectiveness of nutrient delivery and absorption by the leaves. Additionally, the targeted nature of foliar application ensures that the nutrients are applied directly where they are needed, minimizing wastage and optimizing nutrient utilization by the plant (Tombuloglu *et al.* 2020).

The observed superiority of spraying the Nano form on leaves in comparison to the chelated form, for both Fe and Zn treatments, can be attributed to that Nano-sized particles have the advantage of smaller size and high surface area, allowing them to penetrate leaf tissues more effectively. This enables a faster and more efficient absorption of nutrients into the plant, promoting better growth and nutrient utilization. Foliar application, especially using Nano forms, provides a direct route for nutrient uptake through the leaves. The nutrients bypass the root system and are immediately available for plant utilization, resulting in quicker responses to nutrient deficiencies and improved plant health. Nano forms are less prone to runoff and can adhere more effectively to leaf surfaces, reducing the risk of nutrient loss through wash-off or leaching. This results in a higher proportion of the applied nutrients being retained and utilized by the plant (Salem, 2022). Foliar sprays can be finely tuned to target specific plant tissues or growth stages. Nanoparticles can be engineered for precision, directing nutrients to areas where they are needed most, such as young leaves or developing fruits, optimizing nutrient utilization. The Nano form can be designed for controlled nutrient release, providing a sustained supply of Fe and Zn over time. This prolonged availability of nutrients supports consistent plant growth and mitigates the risk of nutrient deficiencies during critical growth stages. Nano-sized particles can be transported more efficiently within leaf tissues, ensuring even distribution of nutrients to various parts of the plant. This leads to balanced growth and better nutrient utilization. The results obtained in this study align with findings from prior research executed by Hossain and Bezbaruah (2021), Zafar *et al.* (2022), Turan *et al.* (2022) and Sun *et al.* (2023), demonstrating consistency and support for the observed trends and outcomes.

The observed interaction effect, particularly the combination of $\mathbf{T_2}$ treatment with $\mathbf{F_2}$ treatment, which resulted in the best performance for spinach plants, can be attributed to several key reasons;

The interaction between the selected $\mathbf{T_2}$ and $\mathbf{F_2}$ treatments likely created a synergistic effect in nutrient uptake. The combination of NPK in both mineral and Nano forms, along with Fe-Nano, may have provided an optimal balance of essential nutrients, promoting robust growth and higher yields. The $\mathbf{T_2}$ treatment, which included both mineral and Nano forms, could have provided a more balanced and sustained supply of nutrients to the plants (Sadati-Valojai *et al.* 2021). This balanced nutrient supply, along with the added Fe-Nano treatment, likely enhanced nutrient availability at critical growth stages. The combined treatment may have optimized nutrient targeting, ensuring that nutrients were directed to the right plant tissues and growth areas. This precise nutrient delivery contributed to enhanced growth parameters and yield. The $\mathbf{T_4}$ treatment (25% recommended dose of NPK as Nano form) in combination with no foliar application ($\mathbf{F_1}$ treatment) resulted in the lowest values. This outcome is likely due to the insufficient nutrient supply from the $\mathbf{T_4}$ treatment and the absence of foliar nutrient application, leading to nutrient deficiency and suboptimal plant growth (Adisa *et al.* 2019).

In summary, the highest values observed in the interaction effect of the $\mathbf{T_2}$ and $\mathbf{F_2}$ treatments are likely a result of synergistic nutrient uptake, balanced nutrient supply, efficient nutrient utilization, controlled release, optimized nutrient targeting, and the prevention of nutrient deficiency. These factors contributed to the superior growth and yield of spinach plants in this specific combination. The results are consistent with the findings obtained by Hossain and Bezbaruah (2021), Jose *et al.* (2021), Gil-Díaz *et al.* (2022) and Salem (2022).

The findings of this study align with existing literature on Nano-fertilizers and spinach cultivation, which suggests that the supplementation of Nano-fertilizers can enhance plant growth and yield compared to conventional fertilizers. The observed superior performance of the T2 treatment, which included a combination of traditional NPK fertilizers and Nano-fertilizers, is consistent with previous research indicating that Nano-fertilizers can improve nutrient uptake efficiency and enhance plant growth parameters (Dhaliwal *et al.* 2021; El-Desouky *et al.* 2021; Ibrahim and Hegab 2022). This improvement may be attributed to the unique properties of Nano-fertilizers, such as their small size, high surface area-to-volume ratio, and targeted nutrient delivery mechanisms, which facilitate better nutrient absorption by plant roots. Additionally, the greater effectiveness of Fe treatments compared to Zn treatments is in line with the known importance of iron in promoting chlorophyll synthesis and photosynthetic activity, crucial for overall plant growth and development (Mahdieh *et al.* 2018; Turan *et al.* 2022).

The observed differences in plant growth and yield between different treatments may be attributed to several potential mechanisms. Firstly, the Nano-form of fertilizers may enhance nutrient availability and uptake by plants due to their ability to penetrate cell walls and deliver nutrients directly to plant cells (Xu *et al.* 2018). Secondly, the controlled-release properties of Nano-fertilizers may ensure a steady supply of nutrients to plants over an extended period, preventing nutrient deficiency and promoting continuous growth (Gil-Díaz *et al.* 2022). Furthermore, the foliar application of Nano-fertilizers may allow for rapid nutrient absorption through the stomata, bypassing potential soil nutrient limitations and increasing nutrient efficiency. Additionally, the chelated forms of Fe and Zn may have exhibited lower effectiveness due to reduced bioavailability compared to Nano-forms, which can readily release nutrients in plant-available forms Abdel-Hakim *et al.* (2023).

Despite the promising results, several limitations and challenges may have influenced the study outcomes. Variability in soil conditions, such as nutrient levels and pH, could have affected nutrient availability and uptake by plants, potentially confounding the results. Environmental factors such as temperature, humidity, and precipitation may have also influenced plant growth and yield, introducing variability across experimental units. Furthermore, the short duration of the study may not fully capture the long-term effects of Nano-fertilizers on soil health, microbial communities, and ecosystem dynamics (Al-Jubouri *et al.* 2023; Abdalla *et al.* 2023).

The findings of this study have significant implications for agricultural practices, suggesting that the integration of Nano-fertilizers with conventional fertilization strategies can enhance crop productivity while reducing the environmental impact of nutrient runoff (Sharaf-Eldin *et al.* 2022). Future research directions may include investigating the optimal dosage, timing, and application methods of Nano-fertilizers for different crops and soil types, as well as assessing their long-term effects on soil fertility, plant-microbe interactions, and ecosystem sustainability. Additionally, studies examining the economic feasibility and scalability of Nano-fertilizers in large-scale agricultural systems are warranted to facilitate their widespread adoption and commercialization (Helaly *et al.* 2021; Shaib and Hany, 2023).

5. Conclusion

The study highlights the effectiveness of supplementing traditional NPK fertilizers with Nano-fertilizers, particularly when combined with foliar applications of Fe and Zn, in enhancing the growth, chemical constituents, and quality parameters of spinach plants. The combination of 75% recommended NPK in traditional bulk form and 15% in Nano form, along with Fe- Nano foliar treatment at a rate of 10mg L^{-1}, consistently resulted in the highest positive impacts across various parameters. These findings suggest the potential for more sustainable and efficient fertilizer practices in spinach cultivation. Therefore, it is recommended that farmers and agricultural practitioners consider adopting this approach to optimize crop performance while minimizing the environmental impact of conventional chemical fertilizers.

Conflicts of interest

The authors have declared that no competing interests exist.

Formatting of funding sources: The research was funded by the personal efforts of the authors.

6. References

Abdalla, Z. F., El-Bassiony, A. E. M., El-Ramady, H. & El-Sawy, S. (2023). Broccoli biofortification using biological nano-and mineral fertilizers of selenium: A comparative study under soil nutrient deficiency stress. Egyptian Journal of Soil Science, 63(1), 57-66.

Abdel-Hakim, S. G., Shehata, A. S., Moghannem, S. A., Qadri, M., El-Ghany, M. F. A., Abdeldaym, E. A., & Darwish, O. S. (2023). Nanoparticulate fertilizers increase nutrient absorption efficiency and agro-physiological properties of lettuce plant. Agronomy, 13(3), 691.

Adisa, I. O., Pullagurala, V. L. R., Peralta-Videa, J. R., Dimkpa, C. O., Elmer, W. H., Gardea-Torresdey, J. L., & White, J. C. (2019). Recent advances in Nano-enabled fertilizers and pesticides: a critical review of mechanisms of action. Environmental Science: Nano, 6(7), 2002-2030.

Al-Jubouri, A. H. A., & Abdulrahman, H. B. A. (2023). Effect of Nano-Fertilizer and Boron on Vegetative Growth Characteristics of Two Hybrids of Cabbage Plant (*Brassica oleracea* var. Capitata L.). In IOP Conference Series: Earth and Environmental Science (Vol. 1158, No. 4, p. 042019). IOP Publishing.

Barker, A. V., & Pilbeam, D. J. (2015). Handbook of plant nutrition. CRC press.

Bhushan, B. (2017). "Introduction to nanotechnology". Springer handbook of nanotechnology, 1-19.

Dewis, J., & Freitas, F. (1970). "Physical and chemical methods of soil and water analysis". FAO soils Bulletin, (10).

Dhaliwal, S. S., Sharma, V., Shukla, A. K., Verma, V., Behera, S. K., Singh, P., ... & Hossain, A. (2021). Comparative efficiency of mineral, chelated and nano forms of zinc and iron for improvement of zinc and iron in chickpea (*Cicer arietinum* L.) through biofortification. Agronomy, 11(12), 2436.

Duncan, D. B. (1955). Multiple range and multiple F tests. Biometrics, 11(1), 1-42.

Elawady, R. A. Elsherpiny, M. A., El-Kafrawy,M (2024). Can foliar application of Titanium and Zeolite Soil Addition Enhance Crisphead Lettuce Production, and Reduce N-Leaching or/and Volatilization? Egyptian Journal of Soil Science, 64(3), 699-714.

El-Desouky, H. S., Islam, K. R., Bergefurd, B., Gao, G., Harker, T., Abd-El-Dayem, H., ... & Zewail, R. M. (2021). Nano iron fertilization significantly increases tomato yield by increasing plants' vegetable growth and photosynthetic efficiency. Journal of Plant Nutrition, 44(11), 1649-1663.

Elemike, E. E., Uzoh, I. M., Onwudiwe, D. C., & Babalola, O. O. (2019). The role of nanotechnology in the fortification of plant nutrients and improvement of crop production. Applied Sciences, 9(3), 499.

Elsherpiny, M. A., & Faiyad, R. M. N. (2023). Effect of titanium as a stimulant of atmospheric nitrogen fixation on faba bean plants. Egyptian Journal of Soil Science, 63(4), 465-474.

Farouk, S., El-Hady, M. A. A., El-Sherpiny, M. A., Hassan, M. M., Alamer, K. H., Al-Robai, S. A. & El-Bauome, H. A. (2023). Effect of dopamine on growth, some biochemical attributes, and the yield of crisphead lettuce under nitrogen deficiency. Horticulturae, 9(8), 945.

Gee, G.W. & J.W. Bauder. (1986). "Particle-size Analysis". pp 383-411 In A. Klute (ed.) Methods of Soil Analysis Part 1. Soil Science Society of America Book Series 5, Madison, Wisconsin, USA.

Gil-Díaz, M., García-Gonzalo, P., Mancho, C., Hernández, L. E., Alonso, J., & Lobo, M. C. (2022). Response of spinach plants to different doses of two commercial nanofertilizers. Scientia Horticulturae, 301, 111143.

Gomez; K. A. & Gomez, A.A (1984). "Statistical Procedures for Agricultural Research". John Wiley and Sons, Inc., New York.pp:680.

Guleria, G., Thakur, S., Shandilya, M., Sharma, S., Thakur, S., & Kalia, S. (2022). Nanotechnology for sustainable agro-food systems: The need and role of nanoparticles in protecting plants and improving crop productivity. Plant Physiology and Biochemistry, 194, 533-549

Hagab, R. H., Kotp, Y. H., & Eissa, D. (2018). Using nanotechnology for enhancing phosphorus fertilizer use efficiency of peanut bean grown in sandy soils. Journal of Advanced Pharmacy Education & Research| Jul-Sep, 8(3), 59-67.

Helaly, A. A., Ashmawi, A. E., Mohammed, A. A., El-Abd, M. T., & Nofal, A. S. (2021). Effect of soil application of nano NPK fertilizers on growth, productivity and quality of Lettuce (*Lactuca sativa*). Al-Azhar Journal of Agricultural Research, 46(1), 91-100.

Hesse, P. R. (1971)." A Text Book of Soil Chemical Analysis". Juan Murry (Publisher) Ltd, London.

Hossain, M. E., & Bezbaruah, A. N. (2021). Nano-and microscale iron for Fe fortification in *Spinacia oleracea*. Nanotechnology for Environmental Engineering, 6, 1-12.

Ibrahim, G. A. (2022). Role of bio and Nano fertilization in increasing growth and productivity of spinach in sandy soil. Egyptian Journal of Desert Research, 72(2), 265-283.

Ibrahim, G. A., & Hegab, R. (2022). Improving yield of Barley using bio and nano fertilizers under saline conditions. Egyptian Journal of Soil Science, 62(1), 41-53.

Jackson, M. L. (1967). "Soil Chemical Analysis". Printice-Hall of India, New Delhi.

Jose, S., Pavaya, R. P., Kumar, J. S., & Malav, J. K. (2021). Influence of different combinations of NPK and micronutrients on nutritional status and quality parameters of sesame under loamy sand of Gujarat. J. Pharm. Innov, 11(11), 1846-1851.

Kahandage, P., Rupasinghe, C., Ariyawansha, K., & Piyathissa, S. (2023). Assessing environmental impacts of chemical fertilizers and organic fertilizers in Sri Lankan paddy fields through life cycle analysis. Journal of Dry Zone Agriculture, 9(1), 45-65.

Litskas, V. D. (2023). Environmental impact assessment for animal waste, organic and synthetic fertilizers. Nitrogen, 4(1), 16-25.

Mahdieh, M., Sangi, M. R., Bamdad, F., & Ghanem, A. (2018). Effect of seed and foliar application of nano-zinc oxide, zinc chelate, and zinc sulphate rates on yield and growth of pinto bean (*Phaseolus vulgaris*) cultivars. Journal of Plant Nutrition, 41(18), 2401-2412.

Mali, S. C., Raj, S., & Trivedi, R. (2020). Nanotechnology a novel approach to enhance crop productivity. Biochemistry and Biophysics Reports, 24, 100821.

Mittal, D., Kaur, G., Singh, P., Yadav, K., & Ali, S. A. (2020). Nanoparticle-based sustainable agriculture and food science: Recent advances and future outlook. Frontiers in Nanotechnology, 2, 579954.

Moreno-Maroto, J. M., & Alonso-Azcarate, J. (2022). Evaluation of the USDA soil texture triangle through Atterberg limits and an alternative classification system. Applied Clay Science, 229, 106689.

Paramo, L. A., Feregrino-Pérez, A. A., Guevara, R., Mendoza, S., & Esquivel, K. (2020). Nanoparticles in agroindustry: Applications, toxicity, challenges, and trends. Nanomaterials, 10(9), 1654.

Peterburgski, A. V. (1968)."Handbook of Agronomic Chemistry". Kolos Puplishing House, Moscow, (in Russian, pp. 29-86).

Porra, R. J., Thompson, W. A. A., & Kriedemann, P. E. (1989). Determination of accurate extinction coefficients and simultaneous equations for assaying chlorophylls a and b extracted with four different solvents: verification of the concentration of chlorophyll standards by atomic absorption spectroscopy. Biochimica et Biophysica Acta (BBA)-Bioenergetics, 975(3), 384-394.

Preetha, P. S., & Balakrishnan, N. (2017). A review of nano fertilizers and their use and functions in soil. Int. J. Curr. Microbiol. Appl. Sci, 6(12), 3117-3133.

Rakhimol, K. R., Thomas, S., Kalarikkal, N., & Jayachandran, K. (2021). Nanotechnology in controlled-release fertilizers. In Controlled Release Fertilizers for Sustainable Agriculture (pp. 169-181). Academic Press.

Richards, L. A. (1954). "Diagnosis and improving of Saline and Alkaline Soils". U. S., Salinity Laboratory Staff. Agric. Handbook, No.60.

Rop, K., Karuku, G. N., Mbui, D., Njomo, N., & Michira, I. (2019). Evaluating the effects of formulated Nano-NPK slow release fertilizer composite on the performance and yield of maize, kale and capsicum. Annals of Agricultural Sciences, 64(1), 9-19.

Sadati-Valojai, S. T., Niknejad, Y., Fallah Amoli, H., & Barari Tari, D. (2021). Response of rice yield and quality to Nano-fertilizers in comparison with conventional fertilizers. Journal of Plant Nutrition, 44(13), 1971-1981.

Salem, H. M. (2022). Efficiency of applying Fe and Zn singly or in various combinations as Nano and non-Nano forms of foliar spray on snap bean (*Phaseolus Vulgaris*). Annals of Agricultural Science, Moshtohor, 60(4), 1255-1258.

Shaib, M. H., & Hany, A. H. E. S. (2023). Efficacy of mineral and nano fertilizers on yield and quality of sugar beet crop cv. karam., (*Beta vulgaris*, L). Egyptian Journal of Soil Science, 63(4), 677-687.

Sharaf-Eldin, M. A., Elsawy, M. B., Eisa, M. Y., El-Ramady, H., Usman, M., & Zia-ur-Rehman, M. (2022). Application of Nano-nitrogen fertilizers to enhance nitrogen efficiency for lettuce growth under different irrigation regimes. Pakistan Journal of Agricultural Sciences, 59(3).

Singh, J. P. (1988). A rapid method for determination of nitrate in soil and plant extracts. Plant and soil, 110, 137-139.

Singh, R. P., Handa, R., & Manchanda, G. (2021). Nanoparticles in sustainable agriculture: An emerging opportunity. Journal of Controlled Release, 329, 1234-1248.

Sun, Y., Mfarrej, M. F. B., Song, X., Ma, J., Min, B., & Chen, F. (2023). New insights in to the ameliorative effects of zinc and iron oxide nanoparticles to arsenic stressed spinach (*Spinacia oleracea* L.). Plant Physiology and Biochemistry, 199, 107715.

Tombuloglu, H., Ercan, I., Alshammari, T., Tombuloglu, G., Slimani, Y., Almessiere, M., & Baykal, A. (2020). Incorporation of micro-nutrients (nickel, copper, zinc, and iron) into plant body through nanoparticles. *Journal of Soil Science and Plant Nutrition*, *20*, 1872-1881.

Turan, M., Ekinci, M., Kul, R., Kocaman, A., Argin, S., Zhirkova, A. M., ... & Yildirim, E. (2022). Foliar Applications of humic substances together with Fe/Nano Fe to increase the iron content and growth parameters of spinach (*Spinacia oleracea* L.). Agronomy, 12(9), 2044.

Walinga, I., Van Der Lee, J. J., Houba, V. J., Van Vark, W., & Novozamsky, I. (2013). Plant analysis manual. Springer Science & Business Media.

Xu, J., Luo, X., Wang, Y., & Feng, Y. (2018). Evaluation of zinc oxide nanoparticles on lettuce (*Lactuca sativa* L.) growth and soil bacterial community. Environmental Science and Pollution Research, 25, 6026-6035.

Zafar, S., Perveen, S., Kamran Khan, M., Shaheen, M. R., Hussain, R., Sarwar, N., ... & Siddiqui, M. H. (2022). Effect of zinc nanoparticles seed priming and foliar application on the growth and physio-biochemical indices of spinach (*Spinacia oleracea* L.) under salt stress. PLoS One, 17(2), e0263194.

Zanini, D. J., Silva, M. H., Aguiar-Oliveira, E., Mazalli, M. R., Kamimura, E. S., & Maldonado, R. R. (2018). Spectrophotometric analysis of vitamin C in different matrices utilizing potassium permanganate. European International Journal of science and technology, 7(1), 70-84.

Zhang, Y. P.; X. W. Chen, Z. X. Zhang, M. Z. Chen, K. Chai and L. X. Wang, (2005): Influences of calcium treatment on the growth characters and shelf lives of Chinese cabbage. Chenese Agric Sci. Bulletin, 21: 223-226, 261.

Zhao, L., Lu, L., Wang, A., Zhang, H., Huang, M., Wu, H., ... & Ji, R. (2020). Nano-biotechnology in agriculture: use of nanomaterials to promote plant growth and stress tolerance. Journal of agricultural and food chemistry, 68(7), 1935-1947.