



## Comparative Efficiency of Nano and Chelated Forms of Iron, Zinc and Manganese for Improvement Yield and Water Use Efficiency of Faba Bean Grown under Drought Stress



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**D**ROUGHT is a highly damaging abiotic stress that affects crops' development, functioning, productivity, and quality. In contemporary farming, nanoparticles are advantageous because of their extensive surface area and enhanced ability to penetrate plant leaves when applied as a spray. Lately, nano-fertilizers have been employed in agriculture to help reduce the negative impacts of drought stress. This study aims to investigate the effects of different forms (nano and chelated) of iron (Fe), zinc (Zn), and manganese (Mn) foliar application, as well as their combinations, on the growth, yield, and water productivity of faba bean plants under different soil moisture levels (100, 80, and 60% of field capacity, FC). The results indicated the best readings of traits studied in the faba bean plant were observed under soil moisture at 100% of FC (control) compared to 60% of FC. On the other hand, results showed that the combined foliar application (FA) of FeZnMn-nanofertilizers (FeZnMn-NFs) to faba bean plants yielded the most favorable growth characteristics and chlorophyll content compared to the untreated plants (control). Also, the FA of FeZnMn-NFs treatment resulted in the highest seed yield and macronutrient (NPK) content in both straw and seed. The seed yield under FeZnMn-NFs treatment ( $21.24 \text{ g pot}^{-1}$ ) was significantly more significant than the control ( $15.47 \text{ g pot}^{-1}$ ). Regarding water use efficiency (WUE), the FeZnMn-NFs treatment achieved the highest WUE for the faba bean ( $2.44 \text{ kg m}^{-3}$ ) compared to the control ( $1.60 \text{ kg m}^{-3}$ ). Conversely, the amount of irrigation water applied (IWA) was lowest with the FeZnMn-NFs treatment ( $8.72 \text{ L pot}^{-1}$ ) compared to the control ( $9.64 \text{ L pot}^{-1}$ ). Concerning the interaction between irrigation levels and foliar spray treatments of faba bean plants, there were no significant differences in seed yield between the 100% irrigation level and the 80% level when foliar application of FeZnMn-NFs. Additionally, nano-fertilizers (NFs) demonstrate greater effectiveness than chelated fertilizers (EDTA), significantly enhancing yield and macronutrient content. Thus, the results highlight the crucial role of NFs in mitigating damage from drought stress, improving growth characteristics, and saving 20% of the amount of IWA for faba bean plants, allowing it to be used elsewhere in agriculture. Consequently, these findings suggest that using NFs of Fe, Zn, and Mn as foliar applications (FA) could be a promising approach to boost the growth parameters, seed yield, and WUE of faba bean plants in arid and semi-arid regions.

**Keywords:** Water deficit; Micronutrients; Nano-fertilizers; Nanotechnology; Water use efficiency; and *Vicia faba*.

### 1. Introduction

The faba bean is a prevalent leguminous crop in Egypt, primarily utilized for human nutrition due to its rich vitamins, minerals, and starch (Fayed et al., 2021; Aboseif et al., 2022; Faiyad & Abd EL-Azeiz, 2024). Additionally, it serves as an alternative protein source (Elsherpiny & Kaney, 2023). This versatile crop is used for food and as fodder and green manure because of its high nitrogen content (Abou Khater et al., 2024). Furthermore, it is an adaptable plant that thrives in various climatic conditions and is resistant to biotic and abiotic stresses (Afreen et al., 2024). Its exceptional ability to fix nitrogen biologically enhances soil fertility, making it a valuable component of cropping systems (Bamber et al., 2024; Gu et al., 2024). Farmers growing faba beans require less nitrogen fertilizer than other crops (El-Badry et al., 2023; Simon-Miquel et al., 2024).

Abiotic stress factors like drought significantly reduce crop productivity (Kopecká et al., 2023; Moustafa et al., 2024a). Drought is a crucial method for reducing irrigation water usage and addressing the water-deficit crisis (Kandhol et al., 2022; Moustafa et al., 2024b), which negatively impacts nutritional quality, yield, and seed growth (Naumann et al., 2018). This stress factor affects approximately 25% of agricultural production (Ahmad et al., 2022), influencing various biochemical, molecular, morphological, ecological, and physiological traits and processes throughout all growth stages (Fathi & Tari, 2016). Climate change, a major global issue, has recently led to more frequent and severe drought conditions (Ali et al., 2022).

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Iron, zinc, and manganese are crucial elements for the growth of many plants, including faba bean (Mahmoud et al., 2022a). Iron (Fe) is a vital micronutrient for all organisms (Gupta et al., 2024). Plants absorb  $\text{Fe}^{2+}$  from the soil, so Fe-deficient soils result in Fe-deficient plants (Wairich et al., 2024). Plants utilize Fe in various physiological processes, such as respiration, chlorophyll biosynthesis, redox reactions, and chlorophyll production (Ahmad et al., 2024). Therefore, enhancing the efficiency of Fe fertilizer use is essential (Zhou et al., 2024). Although using Fe in its traditional form remains popular for boosting plant yields, it often proves ineffective due to poor nutrient-use efficiency (Pudhuvai et al., 2024). Zinc (Zn) is another essential micronutrient plant, and zinc deficiency is widespread in several crops (Hacisalihoglu, 2020). Zinc is necessary for the activity of various enzymes, including isomerases, aldolases, transphosphorylases, dehydrogenases, DNA and RNA polymerases, and is involved in cell division, tryptophan synthesis, photosynthesis, membrane structure maintenance, and acts as a regulatory cofactor in the synthesis of protein (Makhasha et al., 2024). Manganese (Mn), a critical micronutrient, is essential for developing pollen tubes and seeds (Lilay et al., 2024). Its role in these processes significantly impacts crop reproduction dynamics and overall yield (Obeng et al., 2024). For plants to complete their reproductive stages and form seeds and pollen tubes necessary for achieving maximum crop productivity, sufficient Mn levels are required (Pudhuvai et al., 2024). Various micronutrients have been utilized to address plant deficiencies, including synthetic chelates like ethylenediamine tetra-acetic acid (EDTA) for micronutrient enrichment in crops (Madhupriyaa et al., 2024). The response of a specific crop can vary depending on the micronutrient source due to transportation efficiency and differences in absorption within the plant (Dhaliwal et al., 2021). Synthetic micronutrient chelates such as EDTA alleviate micronutrient deficiencies and improve plant health (Almendros et al., 2019). FA, which involves spraying liquid fertilizers directly onto plant leaves, is an effective method for maximizing yield and minimizing losses (Niu et al., 2021). Foliar sprays containing micronutrients like Fe, Zn, and Mn are convenient for field use, highly effective, and elicit a rapid response from plants (Ishfaq et al., 2022). These applications are well-known for enhancing plant nutrient content (Mahmoud et al., 2022a). The use of Zn-EDTA has proven effective in boosting agricultural output under drought conditions (Wang et al., 2017).

Nanotechnology is a promising interdisciplinary research field, offering numerous opportunities in medicine, pharmaceuticals, electronics, and agriculture (Kumar et al., 2024; Singh et al., 2024a). Nanomaterials typically refer to materials sized between 1-100 nm (Saleh, 2020). Their small size and large surface area confer unique optical, physical, and biological properties (Joudeh & Linke, 2022). Recently, extensive studies have explored various nanotechnology applications in agriculture, enhancing agricultural practices (Wahab et al., 2024). Nanotechnology can revolutionize the farming and food industries by improving plant nutrient absorption (Ghorbani et al., 2024; Singh et al., 2024b). A key application in crop production is nano-fertilizers (NFs), which provide nutrients to plants slowly and in a controlled manner, unlike conventional fertilizers (Seleiman et al., 2020). These NFs are more efficient, reducing environmental risks and soil pollution associated with chemical fertilizers and increasing production stability by mitigating biotic and abiotic stresses (Kashyap et al., 2017). Nanomaterials can mimic antioxidative enzymes like catalase, superoxide dismutase, and peroxidase (Jomova et al., 2024). These enzymes and antioxidants constantly scavenge reactive oxygen species (ROS) (Rahman et al., 2024).

Nano-fertilizers (NFs) are a crucial agricultural tool for enhancing crop growth, yield, and quality while also increasing nutrient use efficiency and reducing fertilizer waste and cultivation costs (Easwaran et al., 2024; Sheta et al., 2024a; Taha et al., 2024). These fertilizers can be applied to soil or leaves (Abou-Youseff et al., 2022; Avila-Quezada et al., 2022). FA is beneficial under unfavorable soil and weather conditions (Faridvand et al., 2021), as it allows nutrients to enter the plant system directly, minimizing fertilizer waste (Arora et al., 2024). Consequently, FA of NFs results in higher nutrient use efficiency (NUE) and rapidly responds to crop growth (Muñoz-Márquez et al., 2022). Additionally, NFs improve growth parameters, the rate of photosynthesis, chlorophyll production, and dry matter production, leading to increased translocation and production of photosynthates to the plant's various parts than conventional fertilizers (Seleiman et al., 2020). One benefit of NFs is that they can be applied in smaller quantities than traditional fertilizers (Kumar et al., 2023). With less than 100 nm particle size and a larger surface area, NFs can easily penetrate plant leaves, ensuring higher use efficiency and nutrient availability (Mahanta et al., 2019). Nano-fertilizers can enhance fertilizer efficiency, yield, and quality (Seleiman et al., 2020). Additionally, nano-Zn fertilizers enhance plant drought tolerance by increasing chlorophyll b content, lipids, amino acids, and proteins (Sedghi et al., 2013). Numerous reports suggest that applying nanoparticles boosts plant tolerance to water deficit by promoting growth, facilitating nutrient uptake, and activating antioxidant enzymes (Zhang et al., 2018). The objectives of this study are to evaluate the efficacy of foliar application (FA) of iron (Fe), zinc (Zn), and manganese (Mn) in two forms (nano and chelated) on the growth parameters, seed yield, chlorophyll content, straw and seed nutrient content, and water use efficiency of faba bean plants under different soil moisture contents.

## 2. Materials and Methods

### 2.1. Experimental Site and Soil Properties

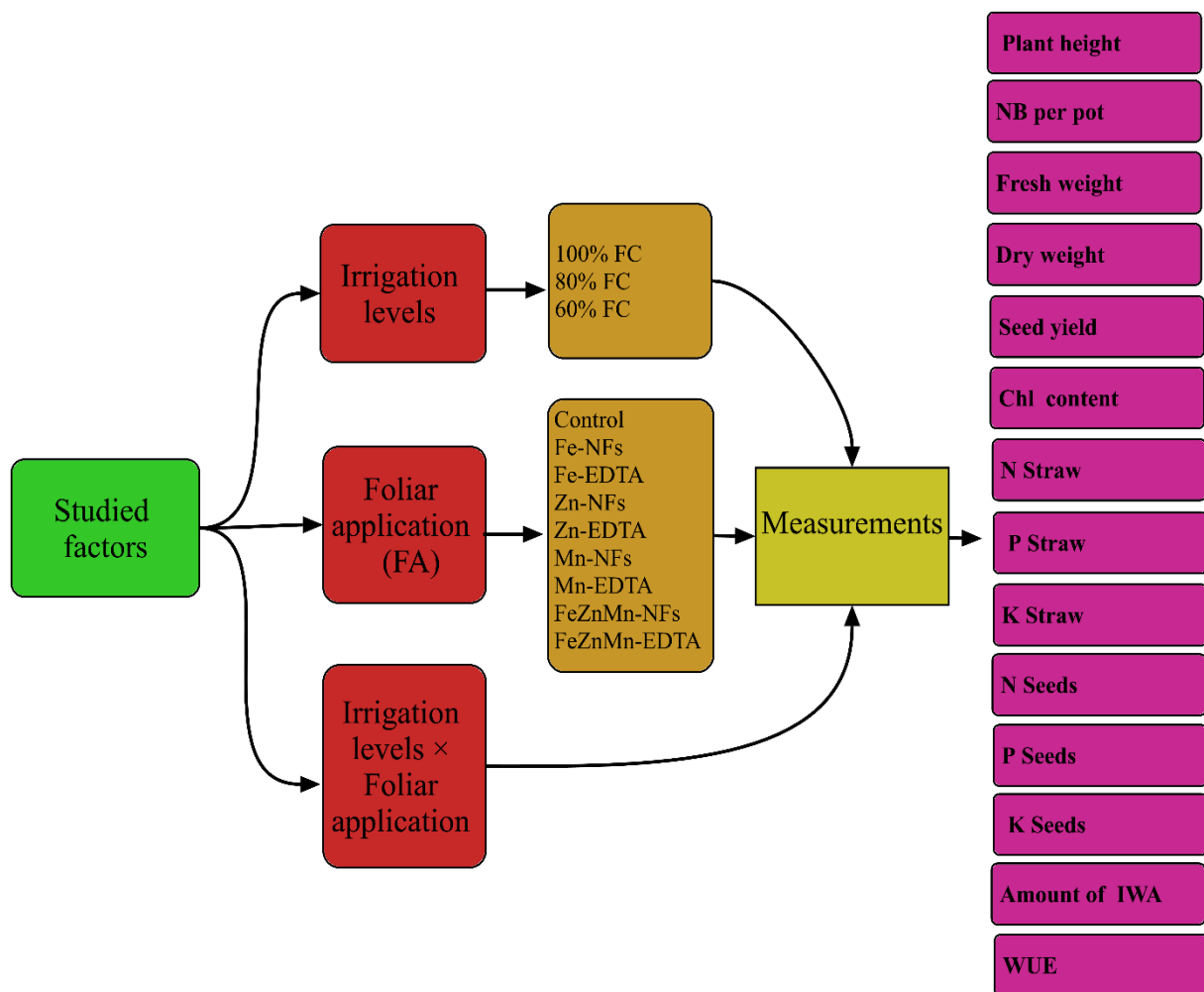
A pot experiment was conducted at the Soils and Water Department, Faculty of Agriculture, Al-Azhar University in Cairo, Egypt, during the winter of 2021/2022. The study aimed to investigate the impact of foliar application (FA) of iron (Fe), zinc (Zn), and manganese (Mn) in both nano and chelated forms on the yield and water use efficiency of faba beans under varying soil moisture levels. The experiment used plastic pots with a capacity of 10 kg, a depth of 35 cm, and a diameter of 30 cm. Soil samples were accumulated from the layer 0.0–30.0cm in the EL-Nubaria region, El Beheira Governorate, Egypt. The physicochemical characteristics of the soil were analyzed, and the results are given in Table 1, following the methods of Dane & Topp (2020) and Sparks et al. (2020).

**Table 1. Some physicochemical characteristics of the studied soil.**

Soil property	Unit	Value	
Particle size distribution:			
Coarse sand		6.33	
Fine sand		76.17	
Silt	(%)	13.87	
Clay		3.63	
Texture class		Loamy sand	
Permanent wilting point		5.89	
Field capacity	(%)	13.67	
Available water		7.78	
ECe (dS m <sup>-1</sup> , soil paste extract)	(dS m <sup>-1</sup> )	3.87	
pH (1:2.5 soil water suspension)		8.52	
Organic matter (OM)	(g kg <sup>-1</sup> )	2.63	
CaCO <sub>3</sub>	(%)	2.29	
Soluble cations (mmolc L <sup>-1</sup> )			
Ca <sup>2+</sup>	8.55	Soluble anions (mmolc L <sup>-1</sup> )	
Mg <sup>2+</sup>	11.45	CO <sub>3</sub> <sup>2-</sup>	0.00
Na <sup>+</sup>	17.85	HCO <sub>3</sub> <sup>-</sup>	2.64
K <sup>+</sup>	0.85	Cl <sup>-</sup>	19.86
		SO <sub>4</sub> <sup>2-</sup>	16.20

### 2.2. Experimental Design and Treatments

The experimental treatments were organized in a randomized complete block design with six replications. The experiment included three irrigation levels of field capacity (FC): 100% FC (full irrigation at 100% of field capacity, control), 80% FC (irrigation at 80% of field capacity), and 60% FC (irrigation at 60% of field capacity). Additionally, foliar application (FA) was performed using each nutrient individually, as well as their combinations, in both chelated (EDTA) and nano (NFs) forms. The treatments were as follows: iron-nano (Fe-NFs), iron-chelated (Fe-EDTA), zinc-nano (Zn-NFs), zinc-chelated (Zn-EDTA), manganese-nano (Mn-NFs), manganese-chelated (Mn-EDTA), iron+zinc+manganese-nano (FeZnMn-NFs), and iron+zinc+manganese-chelated (FeZnMn-EDTA), along with a control treatment (plants treated with redistilled water). The experimental flowchart is illustrated in Fig. 1. These FA treatments were applied at a concentration of 25 mg L<sup>-1</sup>. The experimental pots were placed under a rain shelter to avoid natural precipitation. Fe, Zn, and Mn oxides were utilized as nano-fertilizers (NFs), while their EDTA forms were used as chelated fertilizers. Each foliar application (FA) treatment was administered three times at three-week intervals, beginning forty days after sowing the faba bean seeds. A hand-held sprayer was used to apply the FA treatments to the upper surface of the *V. faba* leaves. The moisture content of the soil was determined using the gravimetric method. For the first 21 days after planting, all pots were watered uniformly. After this period, the irrigation treatments were implemented. Every two days, just before each irrigation, the pots were weighed to measure water loss and add the necessary amount to maintain a consistent field capacity (FC) at 100%, 80%, and 60% FC. The amount of IWA during the growing season was calculated for each FC level.



**Fig. 1.** Experimental flowchart. 100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, 60% FC: irrigation at 60% of field capacity, CT: control, Fe-NFs: iron-nano, Fe-EDTA: iron-chelated, Zn-NFs: zinc-nano, Zn-EDTA: zinc-chelated, Mn-NFs: manganese-nano, Mn-EDTA: manganese-chelated, FeZnMn-NFs: iron+zinc+manganese-nano, FeZnMn-EDTA: iron+zinc+manganese-chelated, NB: number of branches, Chl: chlorophyll, N straw: nitrogen content in straw, P straw: phosphorous content in straw, K straw: potassium content in straw, N seeds: nitrogen content in seeds, P seeds: phosphorous content in seeds, K seeds: potassium content in seeds, IWA: irrigation water applied, and WUE: water use efficiency.

### 2.3. Characterization of Nano-fertilizers

The Fe-NFs and Zn-NFs were supplied by the Nanotechnology and Advanced Materials Central Lab (NAMCL), Agricultural Research Center (ARC) in Giza, Egypt. At the same time, the Mn-NFs were sourced from the NanoFab Technology company in New Maadi, Cairo, Egypt. For transmission electron microscope (TEM) analysis, a drop of the solution was placed on carbon-coated copper grids (CCG) and dried at room temperature, allowing the water to evaporate. A JEOL GEM-1010 TEM captured electron micrographs at 80 kV at the Regional Center for Mycology and Biotechnology of Al-Azhar University, Cairo, Egypt. The TEM analysis was conducted, and the results are shown in Fig. 2. A zeta voltage test was performed using a zeta sizer (Malvern, Nano ZS, UK) at the NAMCL, ARC in Giza, Egypt, to determine the electrical charge and confirm the stability of the Fe-NFs, Zn-NFs, and Mn-NFs solutions. The zeta potential values for Fe-NFs, Zn-NFs, and Mn-NFs were negative, measuring  $-10.7$ ,  $-12.3$ , and  $-11.2$ , respectively, as shown in Fig. 3.

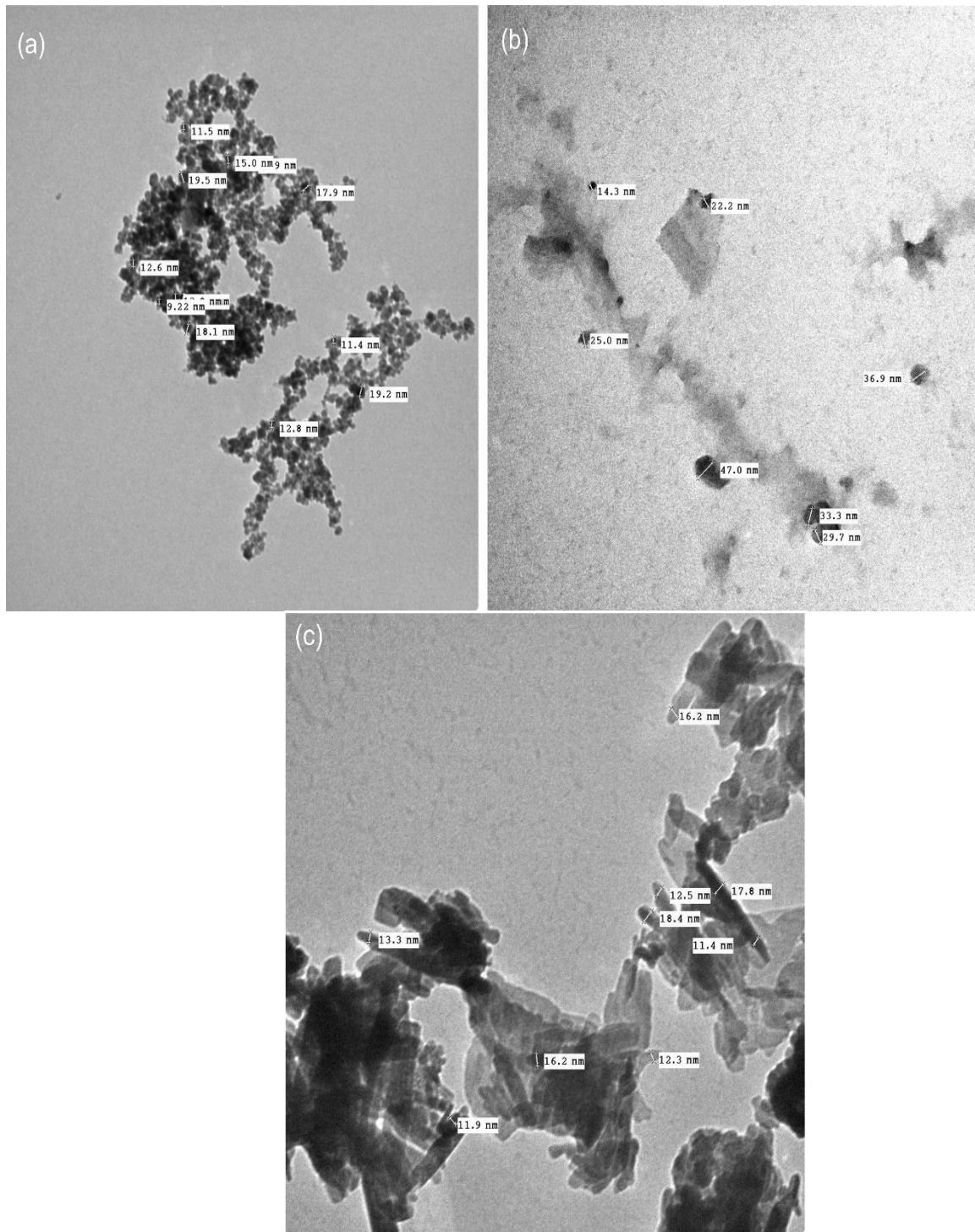
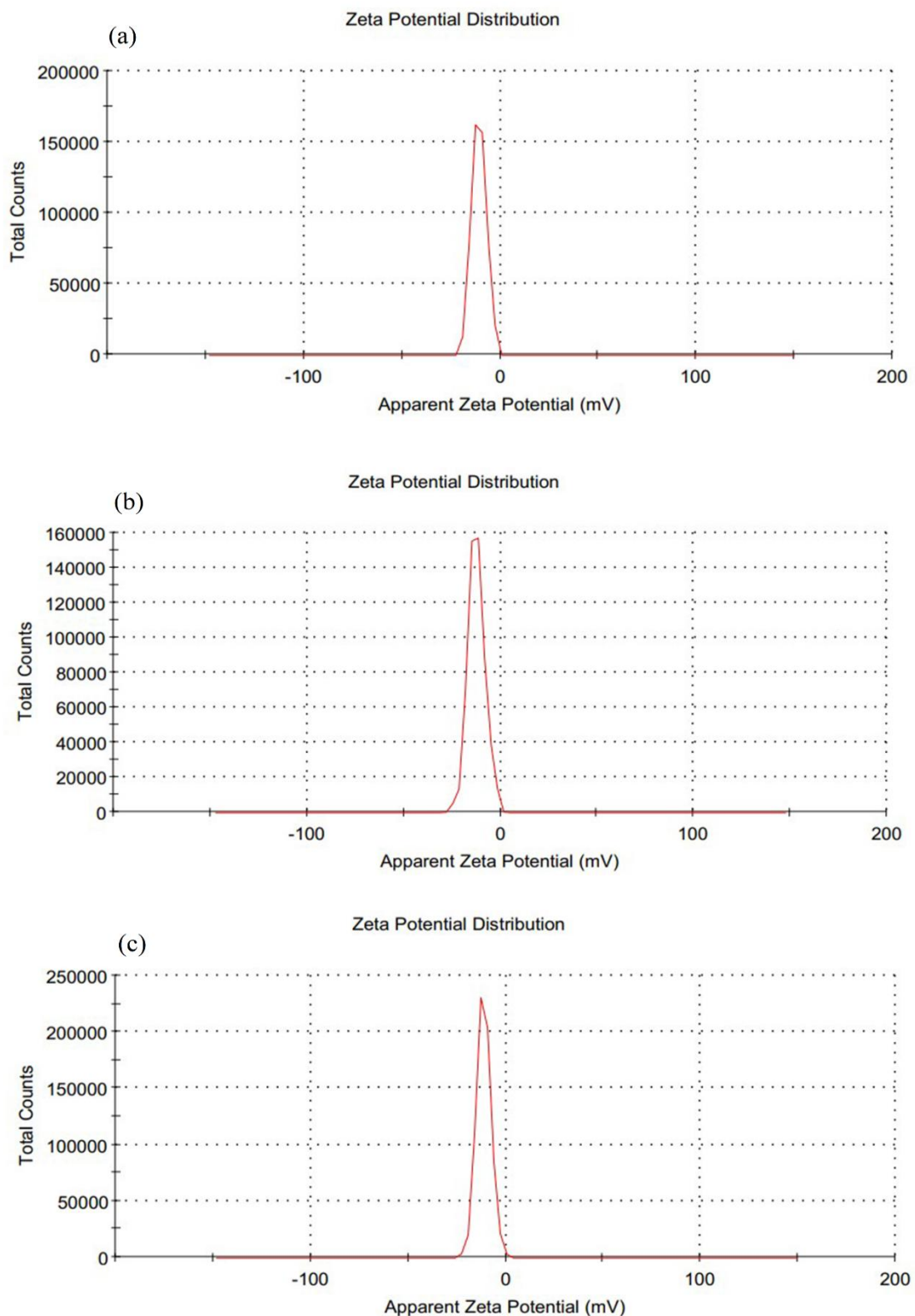


Fig. 2. TEM analysis of Fe (a), Zn (b), and Mn (c) oxide nano-fertilizers.

#### 2.4. Agricultural Practices

Faba bean (*Vicia faba* L.) seeds of the Nubaria 1 variety were provided by the Field Crops Research Institute, ARC in Giza, Egypt. The faba beans were sown on November 17<sup>th</sup> and harvested on April 16<sup>th</sup>. Before sowing, the seeds were inoculated with the *Rhizobium leguminosarum* strain. Five seeds were planted in each pot, and after 21 days, the best three plants were selected per pot. All plants received the recommended organic and mineral fertilizers.



**Fig. 3.** Zeta potential analysis of Fe (a), Zn (b), and Mn (c) oxide nano-fertilizers.

## 2.5. Measurements

After 90 days of the sowing, the traits measured for faba bean plants included plant height (PH, cm) and number of branches per pot (NB pot<sup>-1</sup>). A SPAD chlorophyll meter assessed the relative chlorophyll content (Chl, SPAD). Harvesting was done 150 days after sowing to determine the fresh weight (FW, g pot<sup>-1</sup>), dry weight  
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(DW, g pot<sup>-1</sup>), and seed yield (SY, g pot<sup>-1</sup>). The macronutrient content (NPK) in both the straw and seeds of the faba bean plants was measured according to Walinga et al. (2013). The amount of IWA during the *V. faba* growing season was calculated in liters per pot. Water use efficiency (kg m<sup>-3</sup>) was estimated according to Payero et al. (2008); Abd El-Kader et al. (2023); Sheta et al. (2024b) as follows:

$$\text{WUE} = \frac{\text{Seed yield (kg pot}^{-1}\text{)}}{\text{Irrigation water applied (m}^3\text{ pot}^{-1}\text{)}}$$

## 2.6. Statistical Analysis

The collected dataset of traits was analyzed using univariate statistical methods. Analysis of variance (ANOVA) was conducted for three irrigation levels, nine foliar application treatments, and their interactions to test for significant differences, following the methodology of Gomez & Gomez (1984), using the XLSTAT (Addinsoft, NY, USA) statistical package. Duncan's Multiple Range Test was employed for mean comparisons of mutations. Analysis of the correlation between the studied traits was conveyed using R software.

## 3. Results

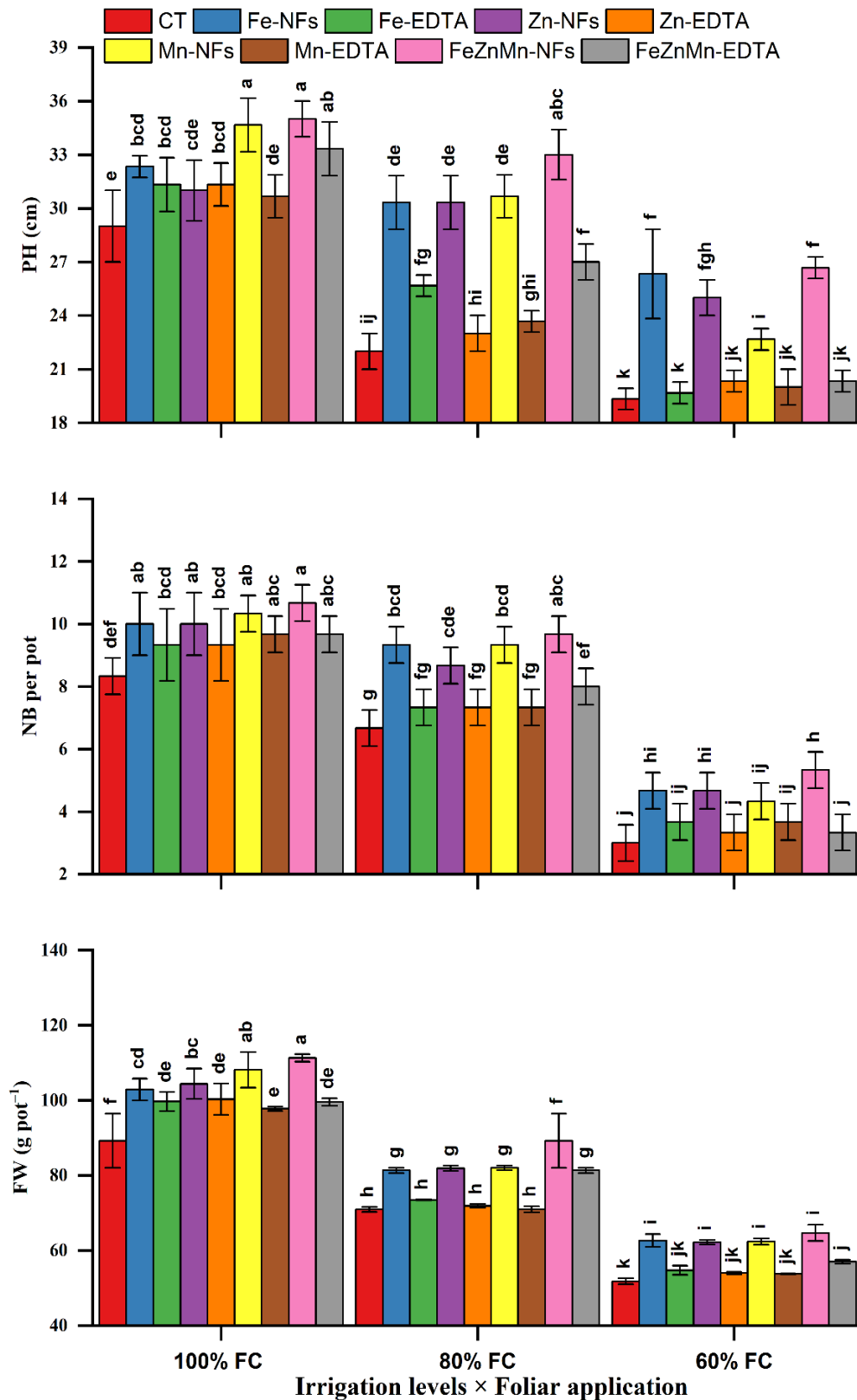
### 3.1. Growth Parameters, Seed Yield, and Chlorophyll Content

As indicated in Table 2, plant parameters such as plant height (PH), number of branches per pot (NB pot<sup>-1</sup>), fresh weight (FW, g pot<sup>-1</sup>), dry weight (DW, g pot<sup>-1</sup>), seed yield (SY, g pot<sup>-1</sup>), and chlorophyll content (Chl, SPAD) significantly increased with full irrigation (100% FC) and the foliar application (FA) of nano-fertilizers (NFs) and/or EDTA, or their combination (Figures 4 and 5). The highest values for PH (32.07 cm), NB (9.70 pot<sup>-1</sup>), FW (101.46 g pot<sup>-1</sup>), DW (58.70 g pot<sup>-1</sup>), SY (21.25 g pot<sup>-1</sup>), and Chl content (49.33 SPAD) were observed in plants irrigated at 100% FC (Table 2). Conversely, the lowest values for PH (22.26 cm), NB (4.00 pot<sup>-1</sup>), FW (58.11 g pot<sup>-1</sup>), DW (35.76 g pot<sup>-1</sup>), SY (14.25 g pot<sup>-1</sup>), and Chl content (37.11 SPAD) were recorded under the 60% FC treatment (Table 2).

**Table 2. Effect of irrigation levels and different forms of Fe, Zn, and Mn as a foliar spray on the growth parameters, seed yield, and chlorophyll content of *V. faba* plants.**

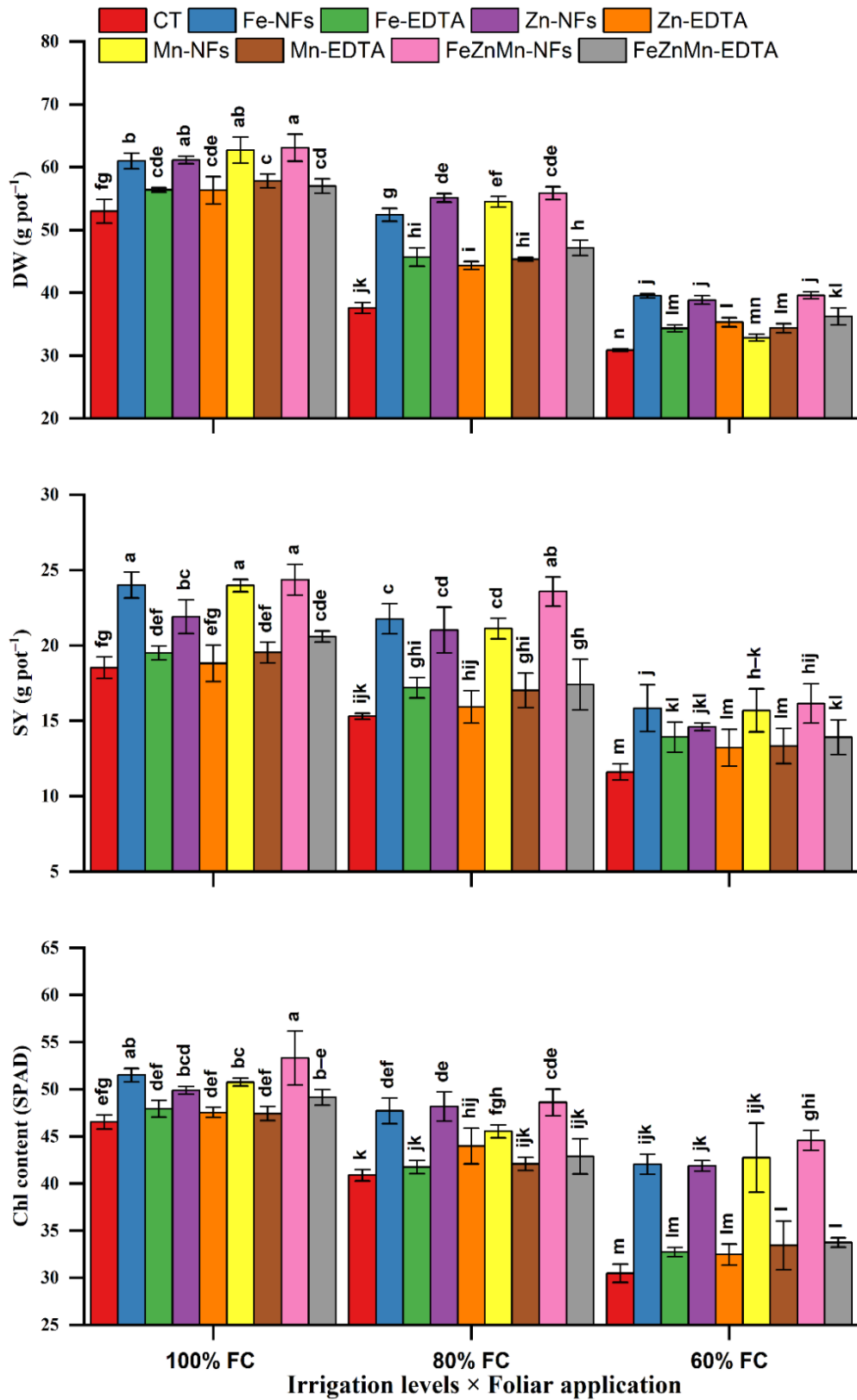
Treatments	PH	NB pot <sup>-1</sup>	FW (g pot <sup>-1</sup> )	DW (g pot <sup>-1</sup> )	SY (g pot <sup>-1</sup> )	Chl (SPAD)
<b>Irrigation levels</b>						
100% FC	32.07±2.2a	9.70±0.95a	101.46±6.84a	58.70±3.54a	21.25±2.38a	49.33±2.34a
80% FC	27.30±3.9b	8.19±1.14b	80.54±12.03b	48.65±5.98b	18.92±2.98b	44.61±3.06b
60% FC	22.26±3.0c	4.00±0.88c	58.11±4.74c	35.76±3.00c	14.25±1.71c	37.11±5.49c
<b>Foliar application (FA)</b>						
CT	24.33±3.9e	6.44±2.19d	72.38±14.46e	40.47±9.87f	15.47±5.24e	39.96±6.18d
Fe-NFs	29.67±3.0ab	8.00±2.60a	82.27±17.53b	50.95±9.40bc	20.65±5.82a	47.92±3.14a
Fe-EDTA	25.56±5.1cd	7.22±1.99bc	74.98±20.80d	45.46±9.58e	16.54±4.07cde	40.79±6.66cd
Zn-NFs	28.78±3.1b	7.78±2.49ab	83.62±17.41b	51.69±10.01ab	19.18±5.53b	46.64±3.75ab
Zn-EDTA	24.89±5.0de	6.67±2.74cd	75.38±20.34d	45.31±9.20e	15.98±4.39de	41.75±6.28c
Mn-NFs	30.11±5.7a	7.78±3.15ab	84.15±20.02b	50.13±13.54c	20.26±5.62a	46.34±3.99b
Mn-EDTA	24.22±5.0e	6.56±2.88cd	74.14±19.22de	45.81±10.17de	16.63±4.69cd	40.96±6.27cd
FeZnMn-NFs	30.78±3.7a	8.22±2.99a	94.89±24.51a	52.71±10.34a	21.24±5.89a	47.98±5.18a
FeZnMn-EDTA	26.56±6.1c	7.00±2.87cd	78.51±19.55c	46.78±9.05d	17.30±4.86c	40.83±8.30cd

PH: plant height, NB pot<sup>-1</sup>: number of branches per pot, FW: fresh weight, DW: dry weight, SY: seed yield, Chl: chlorophyll content, 100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, 60% FC: irrigation at 60% of field capacity, CT: control, Fe-NFs: iron-nano, Fe-EDTA: iron-chelated, Zn-NFs: zinc-nano, Zn-EDTA: zinc-chelated, Mn-NFs: manganese-nano, Mn-EDTA: manganese-chelated, FeZnMn-NFs: iron+zinc+manganese-nano, and FeZnMn-EDTA: iron+zinc+manganese-chelated. Values represent means ±SD. Means within the same column that share the same letter do not differ significantly ( $p < 0.05$  level).



**Fig. 4.** Effect of the interaction between irrigation levels and different forms of Fe, Zn, and Mn as a foliar spray on the plant height (PH), number of branches (NB), and fresh weight (FW) of *V. faba* plants. 100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, and 60% FC: irrigation at 60% of field capacity. Different bars indicate the values of means  $\pm$ SD. Bars with different letters above them indicate a statistically significant difference ( $P < 0.05$  level), whereas bars with the same letters show no significant difference.





**Fig. 5.** Effect of the interaction between irrigation levels and different forms of Fe, Zn, and Mn as a foliar spray on the dry weight (DW), seed yield (SY), and chlorophyll (Chl) content of *V. faba* plants. 100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, and 60% FC: irrigation at 60% of field capacity. Different bars indicate the values of means  $\pm$ SD. Bars with different letters above them indicate a statistically significant difference ( $P < 0.05$  level), whereas bars with the same letters show no significant difference.

The data in Table 2 also revealed that these parameters were significantly enhanced by the FA of Fe, Zn, and Mn in either nanofertilizer (NFs) or EDTA forms. The highest values for PH (30.78 cm), NB per pot (8.22 pot<sup>-1</sup>), FW (94.89 g pot<sup>-1</sup>), DW (52.71 g pot<sup>-1</sup>), SY (21.24 g pot<sup>-1</sup>), and Chl content (47.98 SPAD) were observed in faba bean plants treated with the FA of FeZnMn-NFs. In contrast, the lowest values for PH (24.33 cm), NB (6.44 pot<sup>-1</sup>), FW (72.38 g pot<sup>-1</sup>), DW (40.47 g pot<sup>-1</sup>), SY (15.47 g pot<sup>-1</sup>), and Chl content (39.96 SPAD) were recorded in untreated plants (control).

The results in Figures 4 and 5 indicated the interaction effects of all treatments studied on faba bean.

The highest values for PH (35.00 cm), NB per pot (10.67 pot<sup>-1</sup>), FW (111.28 g pot<sup>-1</sup>), DW (63.07 g pot<sup>-1</sup>), SY (24.36 g pot<sup>-1</sup>), and Chl content (53.30 SPAD) were achieved with the interaction between IWA at 100% FC and FA of FeZnMn-NFs. Conversely, the lowest values for PH (19.33 cm), NB (3.00 pot<sup>-1</sup>), FW (51.80 g pot<sup>-1</sup>), DW (30.87 g pot<sup>-1</sup>), SY (11.60 g pot<sup>-1</sup>), and Chl content (30.47 SPAD) were observed with the interaction between IWA at 60% FC and untreated plants (control).

### 3.2. Straw and Seed Nutrient Content

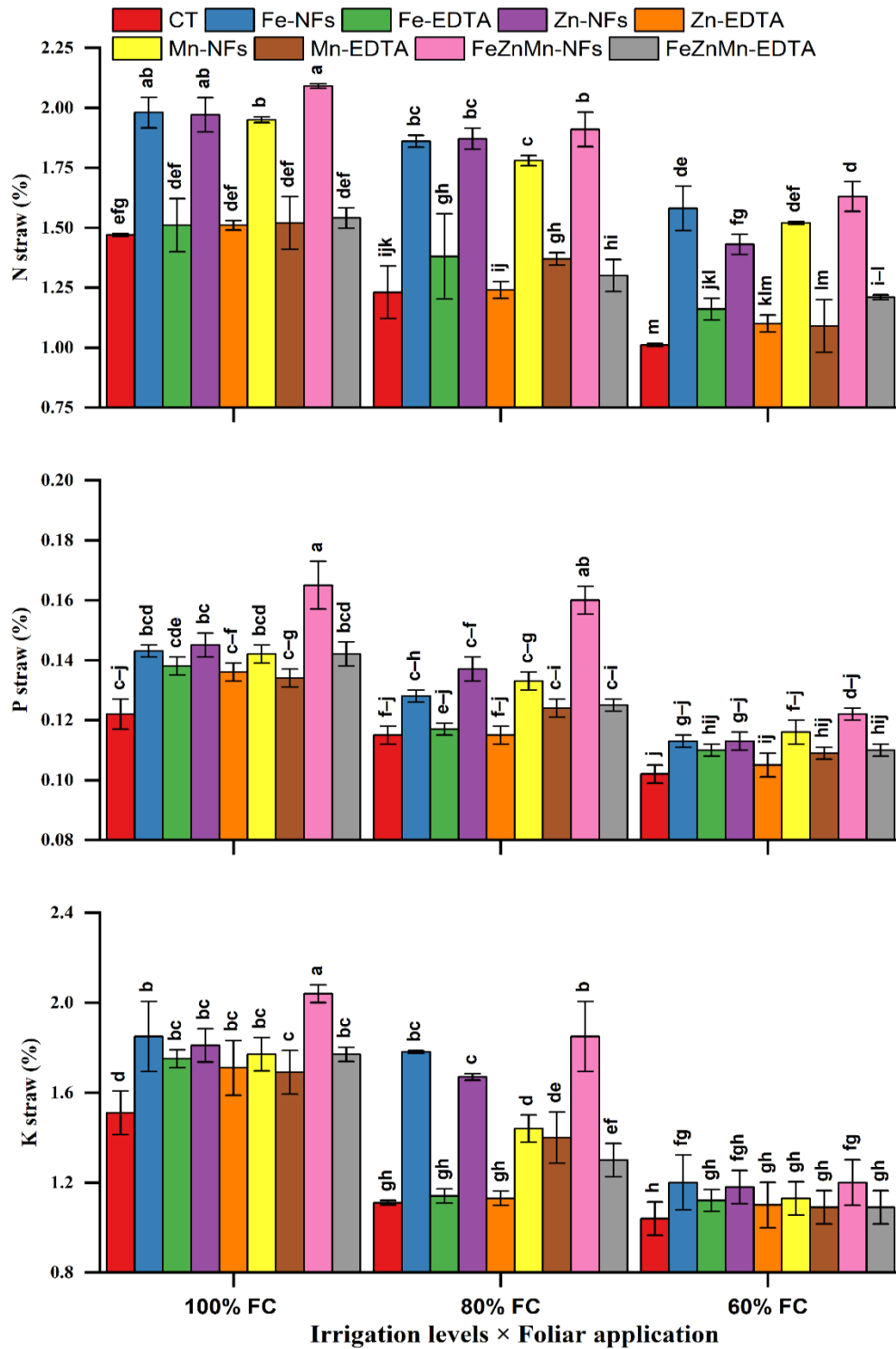
The data in Table 3 indicate that the NPK concentration (%) in straw and seeds of faba bean plants significantly decreased as the irrigation water was reduced from 100% FC to 60% FC. The highest NPK content in straw (1.73%, 0.14%, and 1.76%, respectively) and seeds (2.14%, 0.157%, and 2.14%, respectively) was observed in plants irrigated at 100% FC. Conversely, the lowest NPK content in straw (1.30%, 0.11%, and 1.13%, respectively) and seeds (1.34%, 0.117%, and 1.70%, respectively) were found in plants irrigated at 60% FC.

The findings (Table 3) indicated that FA with nano-fertilizers (NFs) positively influenced the nutrient content in both straw and seeds regarding the impact of FA of Fe, Zn, and Mn in nano and chelated forms. The highest NPK content in straw (1.82%, 0.149%, and 1.75%, respectively) and seeds (2.24%, 0.160%, and 2.39%, respectively) were achieved with the FA of FeZnMn-NFs treatment. In contrast, the lowest NPK content in straw (1.26%, 0.116%, and 1.30%, respectively) and seeds (1.51%, 0.128%, and 1.70%, respectively) were observed in the control treatment (without FA).

**Table 3. Effect of irrigation levels and different forms of Fe, Zn, and Mn as a foliar spray on straw and seed nutrients content of *V. faba* plants.**

Treatments	Nutrient content in straw (%)			Nutrient content in seeds (%)		
	N	P	K	N	P	K
<b>Irrigation levels</b>						
100% FC	1.73±0.258a	0.141±0.015a	1.76±0.151a	2.14±0.228a	0.157±0.012a	2.14±0.221a
80% FC	1.55±0.292b	0.128±0.019b	1.45±0.315b	1.78±0.418b	0.142±0.015b	1.96±0.290b
60% FC	1.30±0.234c	0.111±0.006c	1.13±0.089c	1.34±0.180c	0.117±0.008c	1.70±0.228c
<b>Foliar application (FA)</b>						
CT	1.26±0.184d	0.116±0.006d	1.30±0.196e	1.51±0.535d	0.128±0.021f	1.70±0.395e
Fe-NFs	1.81±0.184a	0.128±0.013bcd	1.45±0.284c	1.88±0.475b	0.149±0.020b	2.02±0.159b
Fe-EDTA	1.75±0.197a	0.130±0.012bc	1.40±0.324cd	1.81±0.298b	0.146±0.21c	1.86±0.179c
Zn-NFs	1.35±0.188bc	0.118±0.016cd	1.34±0.310de	1.64±0.275cd	0.131±0.016e	1.81±0.185cd
Zn-EDTA	1.29±0.181cd	0.118±0.014cd	1.31±0.307e	1.52±0.373d	0.129±0.014f	1.79±0.181d
Mn-NFs	1.37±0.147b	0.131±0.015b	1.55±0.289b	1.74±0.330bc	0.131±0.016e	2.04±0.074b
Mn-EDTA	1.28±0.236cd	0.122±0.011bcd	1.30±0.304e	1.57±0.332d	0.130±0.015ef	1.76±0.189de
FeZnMn-NFs	1.82±0.160a	0.149±0.035a	1.75±0.418a	2.24±0.529a	0.160±0.022a	2.39±0.430a
FeZnMn-EDTA	1.81±0.296a	0.125±0.014bcd	1.61±0.327b	1.87±0.336b	0.143±0.015d	2.02±0.083b

N: Nitrogen, P: phosphorous, K: potassium, 100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, 60% FC: irrigation at 60% of field capacity, CT: control, Fe-NFs: iron-nano, Fe-EDTA: iron-chelated, Zn-NFs: zinc-nano, Zn-EDTA: zinc-chelated, Mn-NFs: manganese-nano, Mn-EDTA: manganese-chelated, FeZnMn-NFs: iron+zinc+manganese-nano and FeZnMn-EDTA: iron+zinc+manganese-chelated. Values represent means ±SD. Means within the same column that share the same letter do not differ significantly ( $p < 0.05$  level).



**Fig. 6.** Effect of the interaction between irrigation levels and different forms of Fe, Zn, and Mn as a foliar spray on nutrient content in the straw of *V. faba* plants. N: Nitrogen, P: phosphorous, K: potassium, 100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, and 60% FC: irrigation at 60% of field capacity. Different bars indicate the values of means  $\pm$ SD. Bars with different letters above them indicate a statistically significant difference ( $P < 0.05$  level), whereas bars with the same letters show no significant difference.

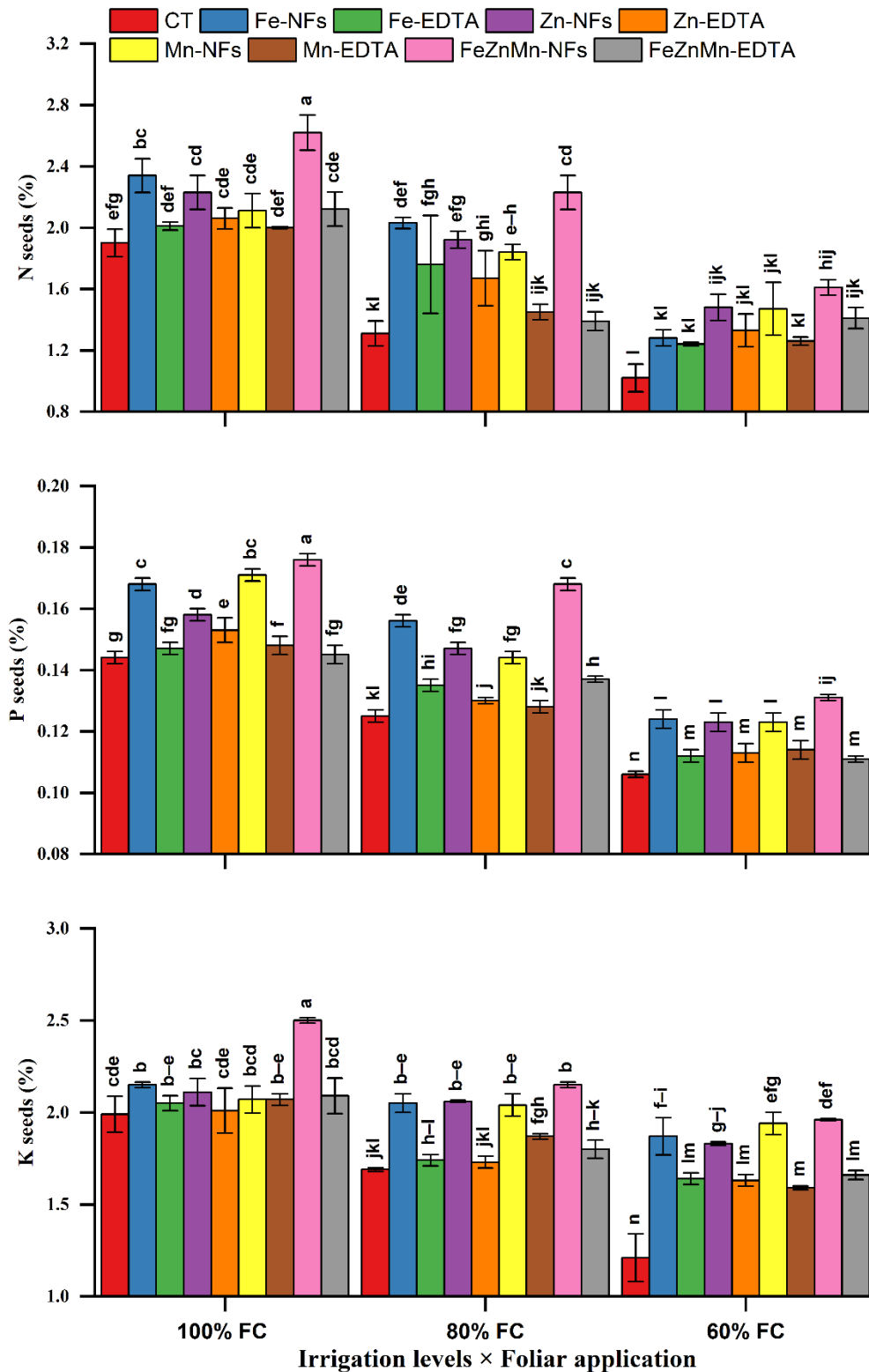


Fig. 7. Effect of the interaction between irrigation levels and different forms of Fe, Zn, and Mn as a foliar spray on nutrient content in the seed of *V. faba* plants. N: Nitrogen, P: phosphorous, K: potassium, 100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, and 60% FC: irrigation at 60% of field capacity. Different bars indicate the values of means  $\pm$ SD. Bars with different letters above them indicate a statistically significant difference ( $P < 0.05$  level), whereas bars with the same letters show no significant difference.

Regarding the interaction between irrigation treatments and foliar spray treatments of faba bean plants, as shown in Figures 6 and 7, the highest NPK content in straw (2.09%, 0.165%, and 2.04%, respectively) and seeds (2.62%, 0.176%, and 2.50%, respectively) was achieved with the combination of IWA at 100% FC and FA of FeZnMn-NFs. Conversely, the lowest NPK content in straw (1.01%, 0.102%, and 1.04%, respectively) and seeds (1.02%, 0.106%, and 1.21%, respectively) were observed with the combination of IWA at 60% FC and without FA (control).

### 3.3. Irrigation Water Applied and Water Use Efficiency

The results in Table 4 demonstrated a significant impact of irrigation levels and FA treatments (NFs and EDTA) on the amount of IWA and WUE of broad bean plants, as well as their interaction (Fig. 8). Increasing the irrigation levels significantly raised the amount of IWA and decreased the WUE. The highest IWA value (13.84 L pot<sup>-1</sup>) was recorded at the 100% FC irrigation treatment, while the lowest IWA value (5.58 L pot<sup>-1</sup>) was observed under the 60% FC treatment (Table 4). Conversely, the highest WUE value (2.55 kg m<sup>-3</sup>) was recorded at the 60% FC irrigation level, whereas the lowest WUE value (1.54 kg m<sup>-3</sup>) was observed at the 100% FC irrigation level.

**Table 4. Effect of irrigation levels and different forms of Fe, Zn, and Mn as a foliar spray on the amount of irrigation water applied (IWA) and water use efficiency (WUE) of *V. faba* plants.**

Treatments	Amount of IWA (L pot <sup>-1</sup> )	WUE (kg m <sup>-3</sup> )
<b>Irrigation levels</b>		
100% FC	13.84±0.66a	1.54±0.19c
80% FC	8.21±0.64b	2.30±0.45b
60% FC	5.58±0.24c	2.55±0.51a
<b>Foliar application (FA)</b>		
CT	9.64±4.81a	1.60±0.36e
Fe-NFs	9.46±4.51ab	2.18±0.80b
Fe-EDTA	9.08±4.45bc	1.82±0.69de
Zn-NFs	9.31±4.56ab	2.06±0.65bc
Zn-EDTA	9.28±4.58ab	1.72±0.62de
Mn-NFs	9.34±4.53ab	2.17±0.62b
Mn-EDTA	9.04±4.41bc	1.84±0.58de
FeZnMn-NFs	8.72±4.18c	2.44±0.79a
FeZnMn-EDTA	9.02±4.26bc	1.92±0.61cd

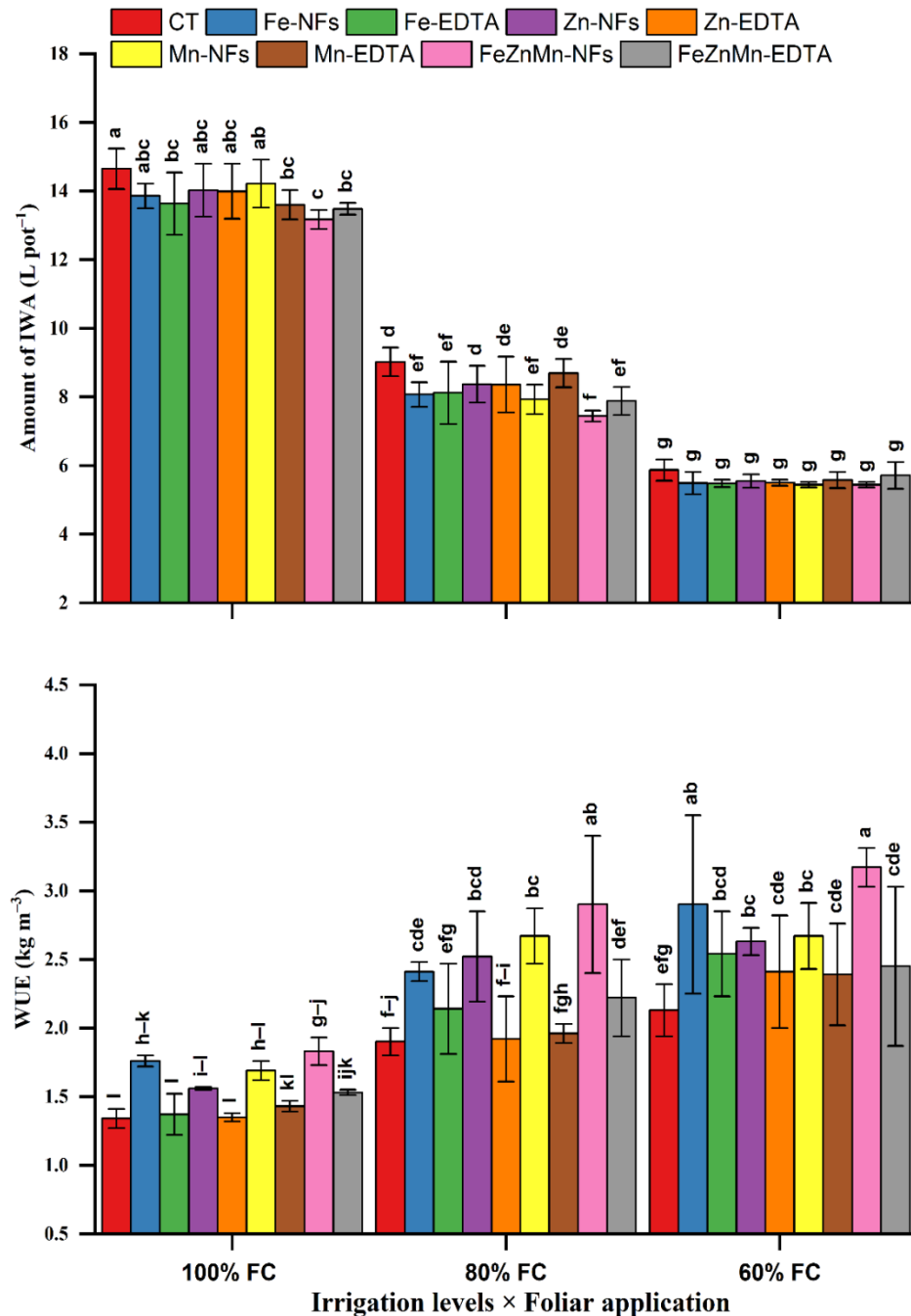
100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, 60% FC: irrigation at 60% of field capacity, CT: control, Fe-NFs: iron-nano, Fe-EDTA: iron-chelated, Zn-NFs: zinc-nano, Zn-EDTA: zinc-chelated, Mn-NFs: manganese-nano, Mn-EDTA: manganese-chelated, FeZnMn-NFs: iron+zinc+manganese-nano and FeZnMn-EDTA: iron+zinc+manganese-chelated. Values represent means ±SD. Means within the same column that share the same letter do not differ significantly ( $p < 0.05$  level).

The data in Table 4 also indicated that the amount of IWA and WUE significantly increased with the FA of NFs and EDTA. The highest IWA value (9.64 L pot<sup>-1</sup>) was observed in the control (without FA), while the lowest IWA value (8.72 L pot<sup>-1</sup>) was recorded with the FA of FeZnMn-NFs treatment. Conversely, the highest WUE value (2.44 kg m<sup>-3</sup>) was achieved with the FeZnMn-NFs treatment, compared to the lowest WUE value (1.60 kg m<sup>-3</sup>) in the control.

Regarding the interaction of all treatments on the amount of IWA WUE values, the data in Fig. 8 showed that the highest IWA value (14.65 L pot<sup>-1</sup>) was observed with the combination of 100% FC irrigation and the control treatment (without FA). In contrast, the lowest IWA value (5.44 L pot<sup>-1</sup>) was recorded using 60% FC irrigation and the FeZnMn-NFs treatment. On the other hand, the highest WUE value (3.17 kg m<sup>-3</sup>) was achieved with the combination of 60% FC irrigation and the FeZnMn-NFs treatment. The lowest WUE value (1.34 kg m<sup>-3</sup>) was compared with the combination of 100% FC irrigation and the control treatment (without FA).

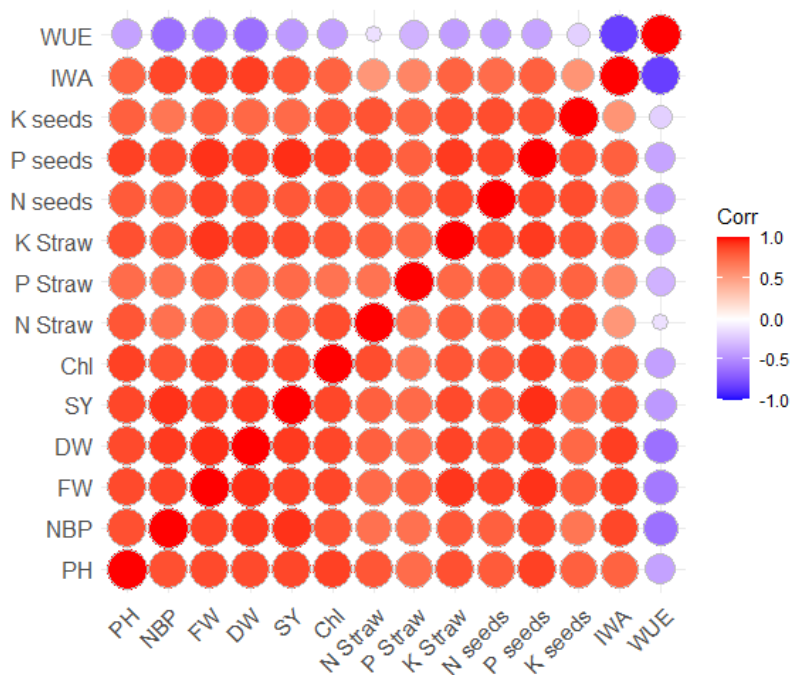
### 3.4. Correlation Analysis

Correlations among measured traits across three irrigation levels and foliar application treatments are presented in Fig. 9. WUE trait showed a negative correlation with all characteristics. In contrast, N straw and K seeds traits showed a non-significant negative correlation ( $p > 0.05$ ) correlation ( $r = -0.134$  and  $-0.197$ ) for N straw and K seeds, respectively. In addition, significant and high negative ( $p < 0.05$ ) correlations ( $r = -0.615^{**}$ ,  $-0.616^{**}$ , and  $-0.829^{**}$ , respectively) were given between WUE and NBP, DW, and IWA, respectively. Moreover, a high positive and significant ( $p < 0.05$ ) correlation was obtained between SY and all traits except WUE. However,



**Fig. 8.** Effect of the interaction between irrigation levels and different forms of Fe, Zn, and Mn as a foliar spray on the amount of IWA and WUE. of *V. faba* plants. IWA: Irrigation water applied, WUE: water use efficiency, 100% FC: irrigation at 100% of field capacity, 80% FC: irrigation at 80% of field capacity, and 60% FC: irrigation at 60% of field capacity. Different bars indicate the values of means  $\pm$ SD. Bars with different letters above them indicate a statistically significant difference ( $P < 0.05$  level), whereas bars with the same letters show no significant difference.

correlation coefficients ranged from  $r = 0.737^{**}$ , given by P straw, to  $r = 0.940^{**}$ , presented by P seeds. On the other hand, IWA traits showed a highly positive and significant ( $p < 0.05$ ) correlation with all traits except WUE. However, a moderate correlation was given by IWA and N straw ( $r = 0.542^{**}$ ) and K seeds ( $r = 0.553^{**}$ ). Correlations were positively correlated with the most significant correlation coefficients between studied traits.



**Fig. 9. Correlation heatmap for studied traits. PH: plant height, NBP: number of branches per pot, FW: fresh weight, DW: dry weight, SY: seed yield, Chl: chlorophyll content, N straw: nitrogen content in straw, P straw: phosphorous content in straw, K straw: potassium content in straw, N seeds: nitrogen content in seeds, P seeds: phosphorous content in seeds, K seeds: potassium content in seeds, IWA: irrigation water applied, and WUE: water use efficiency.**

#### 4. Discussion

The results of this study showed that parameters such as plant height, number of branches, fresh weight, dry weight, seed yield, and chlorophyll content of faba bean significantly increased compared to the control treatment when Fe, Zn, and Mn fertilizers were applied, regardless of the form used. The FA of combined Fe, Zn, and Mn fertilizers resulted in a higher seed yield than single applications due to the excellent supply of these nutrients. Combining nano-fertilizers (FeZnMn-NFs) proved more effective than the chelated fertilizers (FeZnMn-EDTA). This effectiveness is likely due to the enhanced supply of Fe, Zn, and Mn, which promotes chlorophyll synthesis and the photosynthetic apparatus, leading to higher yields. Similar findings, indicating enhanced vegetative growth parameters, could be attributed to increased chlorophyll content, photosynthesis rate, and nutrient uptake, significantly boosting the accumulation of polysaccharides and organic matter in various plant organs (Elfeky et al., 2013). Faba bean responded positively to the foliar application (FA) of micronutrients. The FA of iron (Fe) improved growth parameters, likely due to enhanced protein synthesis and carbohydrate and increased photosynthesis (Chao & Chao, 2022).

Additionally, Fe is essential for synthesizing growth promoters such as auxins, chlorophyll production, nucleic acid metabolism, and seed maturation (Liu et al., 2014). Zinc is vital for auxin production, which regulates growth and development and promotes the formation of auxin compounds like indole acetic acid (El-Boray et al., 2024). Mn activates enzymes involved in photosynthesis and respiration, which are crucial for nitrogen metabolism and absorption (Andresen et al., 2018). Consequently, improvements in these parameters lead to higher seed yields (Hafeez et al., 2022). These findings are consistent with the results reported by El-Gioushy et al. (2021), who found that spraying nano-Fe significantly enhances plant growth parameters and fruit quality compared to chelated and conventional Fe treatments due to increased photosynthesis pigments and nutrient absorption. Nano-fertilizers demonstrated higher content and translocation efficiency than chelated fertilizers

(Pudhuvai *et al.*, 2024). The current results are consistent with previous studies, where applying nano-fertilizers in faba beans resulted in higher yields than chelated fertilizers (Mahmoud *et al.*, 2022a). Similarly, Nadi *et al.* (2013) found that applying Fe-NFs positively impacted faba bean yield and yield parameters. Chlorophyll is a crucial pigment in photosynthesis, absorbing and transferring light energy (Simkin *et al.*, 2022). The chlorophyll content in leaves is a key indicator of the physiological performance of plant photosynthetic tissues, significantly affecting plant photosynthesis (Sharma *et al.*, 2020). The present study showed that faba beans treated with nano-fertilizers (NFs) had superior leaf chlorophyll content. This improvement was partly due to Fe-NFs enhancing the activity of Fe oxygen reductase, which indirectly boosted porphyrin metabolism to produce aminolevulinic acid, a chlorophyll precursor (Li *et al.*, 2018). Similarly, Plaksenkova *et al.* (2019) found that Fe-NFs increased chlorophyll levels in the leaves of *Eruca sativa* compared to the control (CT) treatment.

The foliar application of Fe, Zn, and Mn, individually and in combination, regardless of the form used, resulted in significantly higher NPK content (%) in the straw and seeds of faba bean compared to the control. This increase was attributed to the immediate availability of nutrients, as FA directs nutrients to the leaves. Generally, the higher nutrient content (%) in the straw and seeds of faba bean plants treated with nano-fertilizers (NFs) compared to chelated fertilizers may be due to the properties of NFs, which have a smaller surface area and higher absorption. NFs of nutrients increased the macronutrient content more effectively than chelated forms. The combined application of FeZnMn-NFs positively influenced the NPK content in the straw and faba bean seeds. This enhancement in macronutrient (NPK) content could be attributed to the foliar spraying of different micronutrients, which improved plant nutrient absorption. Nano-fertilizers (NFs) demonstrated more significant potential in increasing NPK content in both straw and seeds than chelated fertilizers, as the single application of NFs was more effective than the single application of chelated fertilizers. Similar findings on the higher effectiveness of NFs compared to chelated fertilizers have been noted by Ponce-García *et al.* (2019). Mahmoud *et al.* (2022a) reported that NFs are more effective than chelated fertilizers in enhancing nutrient content in faba bean plants. The increase in N content with Fe fertilization can be attributed to Fe's crucial role in promoting nodule formation and improving symbiotic associations, thereby boosting nitrogen fixation (NF) (Brear *et al.*, 2013). Abou El-Nasr *et al.* (2015) found that FA with Fe oxide nanoparticles at a concentration of 250 ppm resulted in the highest nitrogen content in pear leaves. Similarly, Askary *et al.* (2017) reported significant increases in Fe, phosphorus (P), and potassium (K) content in the presence of Fe, especially Fe nanoparticles. Consequently, many researchers have concluded that using NFs at optimal concentrations significantly improves crops' vegetative growth, yield, and nutritional status in arid and semi-arid regions and nutrient-poor sandy soils (Abou Tahoun *et al.*, 2022; El-Gioushy *et al.*, 2021).

The current study demonstrated that foliar application (FA) of nano-fertilizers (NFs) led to improved WUE, followed by chelated fertilizers, compared to unfertilized plants (control). The highest amount of IWA was observed in control treatment, followed by chelated fertilizers and plants treated with NFs. This positive trend can be attributed to the ability of NFs containing Fe, Zn, and Mn to mitigate the inhibitory effects of drought stress on plants. Zinc-based micronutrient fertilizers have been shown to alleviate water stress in plants by detoxifying drought-induced free radicals, maintaining cell integrity, and improving water potential. Dimkpa *et al.* (2019) support these findings. Various studies suggest that Zn-based fertilizers are crucial in mitigating plant drought stress by enhancing photosynthetic pigments, reducing lipid peroxidation, and reactive oxygen scavenging substances (Rai-Kalal *et al.*, 2021). Mahmoud *et al.* (2022b) found that an increase in photosynthesis rate will likely improve plants' water use efficiency (WUE). Consequently, this study demonstrated that NFs significantly help reduce drought stress's adverse effects. These conclusions are corroborated by Abou-Sreea *et al.* (2022), Al-Selwey *et al.* (2023), Ghiyasi *et al.* (2023), and Liu *et al.* (2024), who observed that adding NFs to plants under water stress conditions enhanced WUE.

## Conclusions

The findings of this study indicate that Fe, Zn, and Mn nano-fertilizers, owing to their unique structural properties, can be utilized more effectively than their chelated counterparts. Moreover, the combined use of Fe, Zn, and Mn NFs led to a notable increase in yield and nutrient content (%) in the straw and seeds of faba bean. The study also highlighted the crucial role of Fe, Zn, and Mn in enhancing seed yield, as their availability improved faba bean yield regardless of the form in which they were applied. Conversely, the FeZnMn-NFs treatment achieved the highest WUE under deficit irrigation conditions. Additionally, foliar application of NFs positively impacted the plant's ability to withstand water stress. Therefore, the foliar application of Fe, Zn, and Mn nano-fertilizers is particularly noteworthy for improving the productivity and faba bean nutrient content, addressing malnutrition of micronutrients.



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