Effect of Nano-Copper Fertilizer Concentrations on Some Growth Traits of Faba Bean (*Vicia faba* L.) Varieties

Aya Alaa Mousa[1], Ayyub Juma Abdl-Rhmaan Al-Bayaty[2]

[1,2]Colleg of Education for Women/Tikrit University-Iraq

Email: Aya.a.mousa@st.tu.edu.iq

Summary

The study was conducted during the 2025–2026 agricultural season in the Tikrit District – Salah al-Din Governorate to evaluate the effect of foliar spraying with copper oxide nanoparticles at concentrations (50, 100, 150 mg L^{-1}) compared to a control treatment on the plant's mineral content and some yield traits of three faba bean (Vicia faba L.) varieties (Iraqi, New Zealand, and Spanish), using a Randomized Complete Block Design (RCBD) with three replications. The results showed that spraying at 150 mg L^{-1} of nano-copper fertilizer was significantly superior in increasing the content of mineral elements (nitrogen, phosphorus, and potassium), with increase rates of 33.33%, 79.16%, and 7.45%, respectively, compared to the control treatment, demonstrating the efficiency of this concentration in enhancing mineral absorption and accumulation within plant tissues. The local variety also showed significant superiority in nutrient content compared to the New Zealand and Spanish varieties, achieving increase rates of 21.18%, 21.21%, and 8.07%, respectively, reflecting its genetic superiority in absorption efficiency and nutrient utilization. Regarding yield traits, copper oxide nanoparticle treatments contributed to improving the productive performance of the faba bean plant, as the 150 mg L^{-1} concentration led to a significant increase in seed weight and individual plant yield, with increase rates of 21.18% and 40.68%, respectively, compared to the control. The local variety was also superior in these traits, achieving the highest values compared to the other varieties, while the interaction between the local variety and the highest nano-fertilizer concentration showed the best performance in most studied traits, confirming a clear synergistic effect between genetic efficiency and nano-nutrition. In contrast, the trait of number of seeds per pod did not show a significant response,

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indicating its genetic stability. Overall, the study confirms the effectiveness of copper oxide nanoparticles, especially at 150 mg L□¹, in improving mineral nutrition efficiency, this was reflected in the increased nutritional value of the seeds, and increasing faba bean productivity under local environmental conditions.

Study Location: This study was conducted during the 2025-2026 agricultural season at home in the Tikrit District – Salah al-Din Governorate using plastic pots for the period from October 10, 2025, to May 1, 2026, to study the effect of spraying with different concentrations (0, 50, 100, 150 mg/L) of nano-copper fertilizer on the growth of faba bean varieties at two stages: the first stage on December 10, 2025 (two months after planting), and the second stage on January 10, 2026 (three months after planting).

Keywords: Faba bean Vicia faba L., Foliar spraying, Genetic variation, Elemental content, Yield components.

Introduction:

Faba bean (*Vicia faba* L.) is an important legume crop belonging to the Fabaceae family, which includes many important food and economic crops such as chickpeas, lentils, beans, and peas. Faba bean varieties are divided into two main groups: Vicia faba var. major, with large seeds used for human consumption, and Vicia faba var. minor, with small seeds used for animal feed (1). Faba bean is characterized by its high nutritional value due to its high content of plant protein, carbohydrates, and dietary fiber, in addition to containing important minerals and vitamins such as calcium, iron, potassium, and vitamins A and C, making it an important crop for food security and nutritional health (2; 3). It also has a high capacity to fix atmospheric nitrogen through its symbiotic relationship with *Rhizobium bacteria, contributing to improved soil fertility and reduced need for chemical fertilizers (4). Faba bean productivity is influenced by multiple genetic, environmental, and management factors. Choosing the variety suitable for local environmental conditions is among the most important factors affecting growth improvement and yield increase. Varieties differ in their morphological and physiological traits due to genetic variation, making the development of new high-yielding varieties with better adaptability essential for improving production and reducing agricultural costs (1; 5). In recent years, interest has increased in using organic, bio-, and nano-fertilizers as alternatives or supplements to traditional chemical fertilizers due to their role in improving soil properties, increasing nutrient absorption efficiency, reducing environmental pollution, as well as containing active organic compounds that contribute to improving plant growth and productivity (6; 7). With advances in agricultural technologies, nanotechnology has emerged as a modern method to improve the efficiency of nutrient use, as nano-fertilizers are characterized by their tiny particle size and increased absorption efficiency within the plant compared to traditional fertilizers (8). Copper is an essential micronutrient for plant growth due to its important role in photosynthesis, cellular respiration, and activation of many enzymes, which directly reflects on plant growth and productive efficiency (9; 10). Therefore, the importance of studying the effect of nano-copper fertilizers on the growth and productivity of different faba bean varieties has emerged, aiming to determine the best concentrations that contribute to improving vegetative, physiological, and yield traits of the plant while reducing the environmental impacts of traditional fertilizers.

Materials and Methods:

This study was conducted during the 2025–2026 agricultural season in the Tikrit District, Salah al-Din Governorate, using plastic pots 30 cm in diameter and 40 cm in height, for the period extending from (October 10, 2025, to May 1, 2026). The study aimed to evaluate the effect of foliar spraying with different concentrations of nano-copper fertilizer (0, 50, 100, and 150 mg/L) on the growth of different faba bean varieties, added in two-time stages for measurement; the first stage on December 10, 2025 (two months after planting), and the second stage on January 10, 2026 (three months after planting). The experiment was conducted according to a Completely Randomized Design (CRD), the treatment repeated three times, and means were compared using Duncan's Multiple Range Test at a significance level of 0.05. Three faba bean varieties were used: the Iraqi (local) variety, the New Zealand variety (Aguadulce), and the Spanish variety (Luz de Otono). Thus, the number of treatments was 12 treatments as shown in Table (1), repeated three times, bringing the total number of experimental units to 36 units. Nano-copper concentrations were coded with the symbol (N), where N0 represents the control treatment (N0) (without addition), N1 at 50 mg/L, N2 at 100 mg/L, and N3 at 150 mg/L, applied as foliar spray to the plants. Varieties were coded with the symbol (V), where V1 represents the Iraqi (local) variety, V2 the New Zealand variety, and V3 the Spanish variety. Irrigation was carried out according to plant needs, and three seeds were planted in each pot to ensure germination uniformity.

Studied Traits

First: Elemental Content in Vegetative Parts: Root and shoot samples were dried in an electric oven at 70°C until constant weight, then ground and stored in airtight containers. Wet digestion was performed using sulfuric and perchloric acids, then extracts were prepared after topping up with distilled water. After that, concentrations of nitrogen, phosphorus, and potassium were estimated in the resulting solution (11).

1. **Plant Nitrogen Content:** Total nitrogen in plant tissues was estimated after digesting the sample with sulfuric and perchloric acids and heating until clear, then distillation was performed using a micro-Kjeldahl apparatus and ammonia was titrated with sulfuric acid, and the percentage was calculated according to the standard equation (12).

N% = (Volume of H2SO4 consumed Normality 14 Dilution factor) / (Sample weight × 1000) 100`

2. Plant Phosphorus Content: Phosphorus was estimated by colorimetric method using a spectrophotometer at a wavelength of 620 nm, depending on the light absorption intensity of the resulting solution (13).

3. Plant Potassium Content: Potassium was estimated in the digested samples

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using an atomic absorption spectrometer at a wavelength of 766.5 nm, after calibration with a standard potassium curve according to the method of (14).

Second: Yield Components and Productivity

1. Number of Pods Plant^{-1}: This trait is calculated by counting all pods formed on a single plant at maturity stage, then the arithmetic mean is extracted for each experimental unit to obtain the number of pods per plant. It is considered an important indicator of productivity and varietal response to agricultural treatments (15).

2. Number of Seeds Pod^{-1}: This trait is calculated by dividing the average number of seeds per plant by the average number of pods per plant to obtain the average number of seeds per pod for each treatment. This trait is considered an important indicator of production quality, reflecting fertilization efficiency, seed formation, and plant response to environmental conditions and agricultural treatments (16).

3. Single Seed Weight (g): This trait is calculated by weighing the total seeds produced from a single plant after harvest and mixing them homogeneously, then dividing the total weight by the number of seeds to obtain the average weight of a single seed. This trait is used as an important indicator of yield quality, efficiency of photosynthetic translocation to seeds, and plant response to agricultural treatments (4).

4. Seed Yield Plant^{-1} (g): This trait is calculated by estimating the yield of a single plant depending on yield components, by multiplying the number of pods per plant × average number of seeds per pod × average weight of a single seed. This indicator reflects the total productive efficiency of the plant and the integration of yield traits (16).

Statistical Analysis

After collection, the data were subjected to statistical analysis using the Minitab program, and differences between means were compared using Duncan's Multiple Range Test at a probability level of 0.05 to determine significant differences between treatments.

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Results and Discussion

First: Elemental Content in Vegetative Parts

1. Plant Nitrogen Content: Figure (1) shows that different concentrations of nano-copper fertilizer had a significant effect on the percentage of nitrogen in the vegetative parts, as spraying with 150 mg L^{-1} resulted in the highest percentage reaching 2.56%, with an increase rate of 33.33% compared to the control treatment, which gave the lowest percentage of 1.92%. The local variety was superior in its nitrogen content, achieving the highest percentage of 2.46% with an increase rate of 21.18% compared to the Spanish variety, which gave the lowest nitrogen percentage of 2.03%. The binary interaction treatment between the local variety and 150 mg L^{-1} of copper oxide nanoparticles achieved the highest nitrogen percentage in the vegetative parts, reaching 2.98%, with an increase rate of 102.72% compared to the interaction between the Spanish variety and zero concentration of copper oxide nanoparticles, which gave the lowest percentage of 1.47%. This behavior is physiologically attributed to the unique properties of nano-particles (Nano-Cu) in terms of enormous surface area and rapid permeability across cellular membranes, which activated oxidative enzymes and raised the activity of the photosynthesis process. This integrative nutritional support interacted synergistically with the genetic advantage of the local variety, which possesses a root system with high efficiency in nutrient absorption and transport, as well as the local variety's adaptation to the hot, dry Iraqi environment (5).

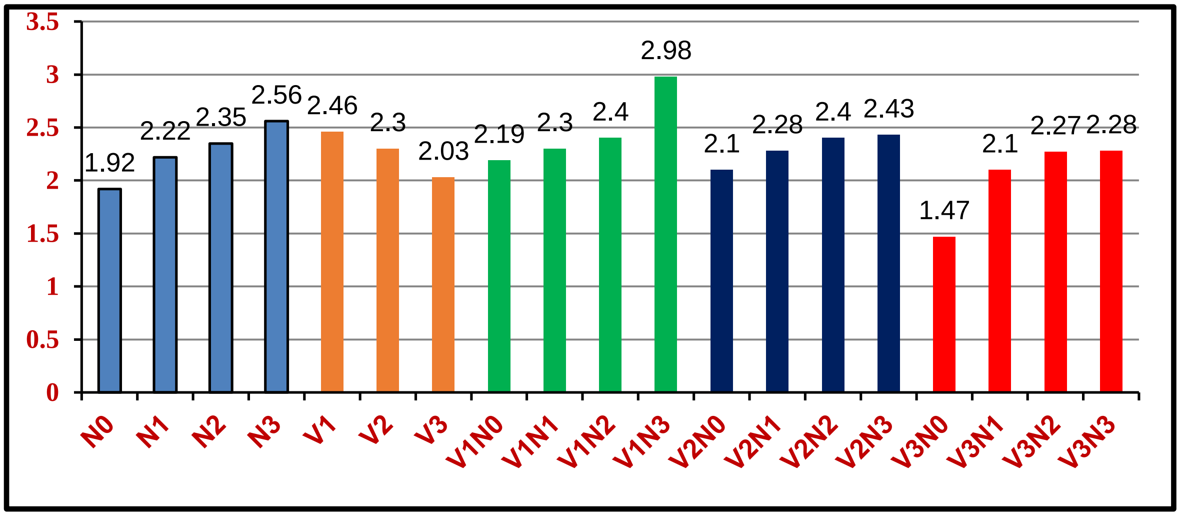

Figure (1). Effect of nano-copper spraying and varieties on plant nitrogen content.

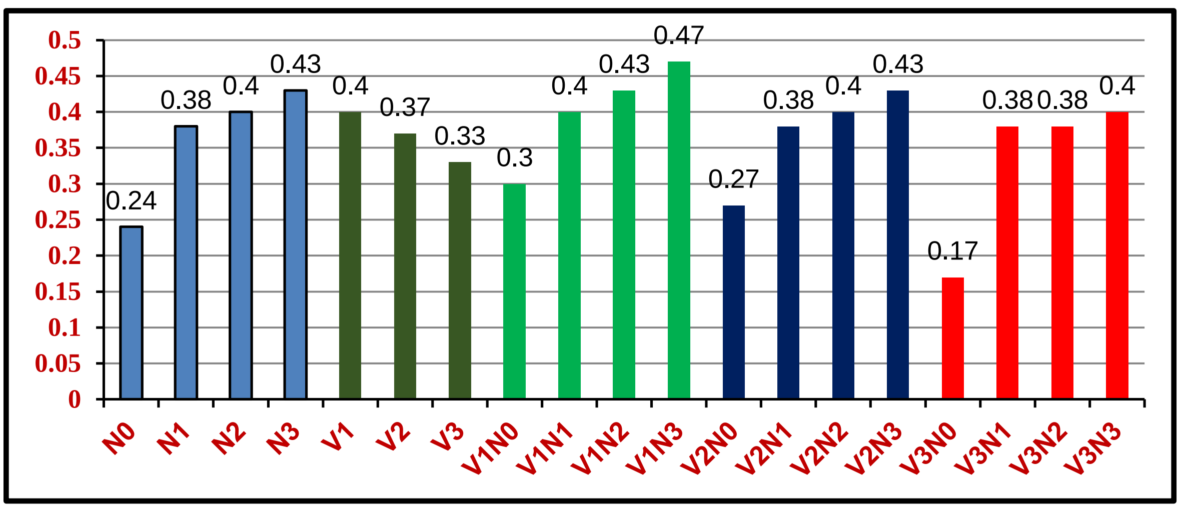

Figure (2). Effect of nano-copper spraying and varieties on plant phosphorus content.

Plant Phosphorus Content: Figure (2) shows that the different concentrations of nano-copper fertilizer had a significant effect on the percentage of phosphorus in the vegetative parts, as spraying with 150 mg L^{-1} achieved the highest percentage of 0.43%, with an increase rate of 79.16% compared to the control treatment, which gave the lowest percentage of 0.24%. The local variety was

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also superior in its phosphorus content, achieving the highest percentage of 0.40% with an increase rate of 21.21% compared to the Spanish variety, which gave the lowest percentage of 0.33%. The binary interaction treatment between the local variety and 150 mg L^{-1} of copper oxide nanoparticles achieved the highest phosphorus percentage in the vegetative parts, reaching 0.47%, with an increase rate of 176.47% compared to the interaction between the Spanish variety and the zero concentration of copper oxide nanoparticles, which gave the lowest percentage of 0.17%. This physiological superiority is attributed to the vital role of copper nanoparticles in increasing the efficiency of root absorption of nutrients, improving their permeability and transport within plant tissues, as well as their contribution to activating many enzymes associated with metabolic and photosynthetic processes, which led to an increased efficiency of phosphorus assimilation within the plant. The superiority of the local variety is also due to its possessing higher genetic potential and distinctive root efficiency in absorbing and utilizing nutrients, in addition to its better adaptation to local environmental conditions, which was clearly reflected in its higher phosphorus content (16).

3. Plant Potassium Content: Figure (3) shows that the different concentrations of nano-copper fertilizer had a significant effect on the percentage of potassium in the vegetative parts, as spraying with 150 mg L^{-1} achieved the highest percentage of 1.73%, with an increase rate of 7.45% compared to the control treatment, which gave the lowest percentage of 1.61%. The local variety was also superior in its potassium content, achieving the highest percentage of 1.74% with an increase rate of 8.07% compared to the Spanish variety, which gave the lowest percentage of 1.61%. The binary interaction treatment between the local variety and 150 mg L^{-1} of copper oxide nanoparticles achieved the highest potassium percentage in the vegetative parts, reaching 1.81%, with an increase rate of 15.29% compared to the interaction between the Spanish variety and the zero concentration of copper oxide nanoparticles, which gave the lowest percentage of 1.57%. This physiological superiority is attributed to the effective role of copper nanoparticles in improving the efficiency of vital processes within the plant, especially increasing cell membrane permeability and activating enzyme systems associated with the absorption and transport of mineral elements, which led to enhanced potassium absorption efficiency and its accumulation in plant tissues. Nano-copper also contributed to raising photosynthetic efficiency and regulating osmotic pressure within cells, which was positively reflected in increased plant mineral content. The superiority of the local variety is due to its possessing more efficient genetic and root traits for nutrient absorption and utilization, as well as its high ability to adapt to local environmental conditions, which enabled it to achieve the highest potassium content compared to the other varieties (15).

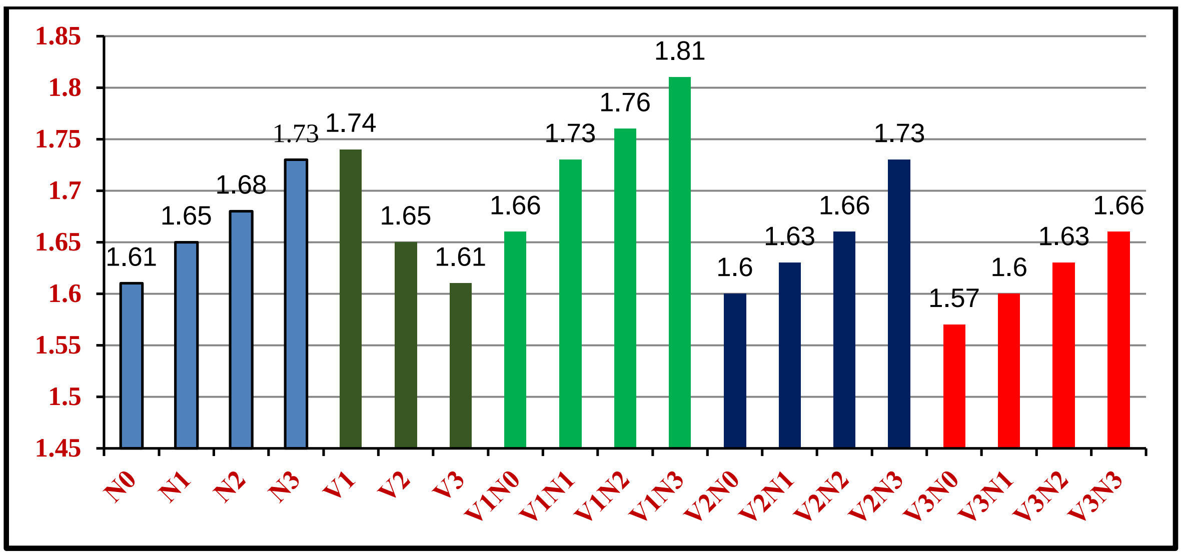

Figure (3). Effect of nano-copper spraying and varieties on plant potassium content.

Third: Yield Components and Productivity

1. Number of Pods Plant□¹: The results in Table (1) show that copper oxide nanoparticle treatments did not show a significant effect on the number of pods for the New Zealand and Spanish varieties, while they showed a significant effect only on the local variety, indicating variable genetic responses among varieties to the nano-treatment. The Spanish

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variety was superior in giving the highest average number of pods plant⁻¹, reaching 6.917 pods, with an increase rate of 29.70% compared to the local variety, which gave the lowest value of 5.333 pods, while the New Zealand variety recorded an intermediate value of 6.833 pods. The results also indicate that treatment with different concentrations of nano-copper fertilizer did not have a significant effect on the number of pods; however, they showed numerical superiority for the 150 mg/L concentration, which gave the highest average number of pods plant⁻¹, reaching 6.667 pods, with an increase rate of 7.55% compared to the control treatment (0 mg/L), which recorded the lowest average of 6.199 pods. This limited response variability is attributed to the fact that the number of pods is a quantitative trait highly correlated with genetic factors rather than being a trait directly affected by fertilizer treatments, making its response to nano-nutrition less pronounced compared to other physiological traits. The relative superiority of some varieties, especially the Spanish one, can be explained by their possessing higher genetic efficiency in flower formation and their conversion into pods, in addition to the efficiency in regulating the hormonal processes responsible for flower setting and development (15).

Table (1). Effect of copper oxide nanoparticles on the number of pods in faba bean plants.

Nitrogen / Varieties	N0	N1	N2	N3	V Mean
V1	4.333 d	5.333 c	5.667 bc	6.000 b	5.333 b
V2	6.667 a	6.667 a	7.000 a	7.000 a	6.833 a
V3	6.667 a	7.000 a	7.000 a	7.000 a	6.917 a
Mean N	6.199 a	6.333 a	6.556 a	6.667 a	

2. Number of Seeds Pod⁻¹: The results in Table (2) show no significant differences between the different concentrations of copper oxide nano-fertilizer in the trait of number of seeds per pod, as all treatments maintained a constant average of 5 seeds pod^{-1}. The varieties also showed no significant differences in this trait, as all varieties recorded the same average of 5 seeds pod^{-1}, indicating the stability of the trait and its lack of influence by the studied factors. Furthermore, the interaction between varieties and different concentrations of copper oxide nanoparticles showed no significant effect, as all interaction treatments maintained the same value of 5 seeds pod^{-1}. These results indicate that the trait of number of seeds per pod is a relatively stable genetic trait that is subject more to genetic control than to the influence of environmental factors or fertilizer treatments. This physiological stability is attributed to the fact that seed formation within pods is associated with a precise genetic regulation of fertilization processes, cell division, and the distribution of nutrients within the pods, making this trait less responsive to changes resulting from the addition of nano-nutrients. Additionally, the favorable environmental conditions during the flowering and pod-setting period may have contributed to maintaining the stability of this trait and the absence of significant variations among different treatments (8).

Table (2). Effect of copper oxide nanoparticles on the number of seeds per pod.

N / V	N0	N1	N2	N3	Mean V
V1	5 a	5 a	5 a	5 a	5 a
V2	5 a	5 a	5 a	5 a	5 a
V3	5 a	5 a	5 a	5 a	5 a
Mean N	5 a	5 a	5 a	5 a	5 a

3. Single Seed Weight (g): Table (3) shows that different concentrations of nano-copper fertilizer had a significant effect on seed weight, as spraying with 150 mg L^{-1} achieved the highest average seed weight of

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0.904 g, with an increase rate of 21.18% compared to the control treatment, which gave the lowest average of 0.746 g. The varieties also showed a significant effect on this trait, as the local variety was superior by giving the highest average seed weight of 1.116 g, with an increase rate of 26.10% compared to the Spanish variety, which gave the lowest average of 0.678 g, while the New Zealand variety recorded an average of 0.698 g. The binary interaction treatment between the local variety and 150 mg L^{-1} of copper oxide nanoparticles achieved the highest seed weight of 1.193 g, with an increase rate of 98.83% compared to the interaction between the Spanish variety and the zero concentration of copper oxide nanoparticles, which gave the lowest seed weight of 0.600 g. This physiological superiority is attributed to the vital role of copper nanoparticles in improving the efficiency of metabolic processes within the plant, especially activating enzymes associated with photosynthesis and the translocation of metabolic products toward the seeds, which contributed to increased accumulation of carbohydrates and proteins within them and increased their weight. Nano-copper also helped to raise nutrient absorption efficiency and improve the physiological status of the plant, which was positively reflected on seed filling and development. The superiority of the local variety is due to its possessing higher genetic capacity to utilize nutrients and convert them into dry matter stored in the seeds, as well as its better adaptation to local environmental conditions compared to the other varieties (11).

Table (3). Effect of copper oxide nanoparticles on seed weight (g).

N / V	N0	N1	N2	N3	Mean V
V1	1.017 d	1.110 c	1.143 b	1.193 a	1.116 a
V2	0.620 k	0.677 i	0.730 g	0.763 e	0.698 b
V3	0.600 l	0.650 j	0.707 h	0.757 f	0.678 b
Mean N	0.746 a	0.812 a	0.860 a	0.904 a	

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4. Seed Yield Plant^{-1} (g): Table (4) shows that different concentrations of nano-copper fertilizer had a significant effect on seed yield per plant, as spraying with 150 mg L^{-1} achieved the highest average yield of 29.670 g, with an increase rate of 40.68% compared to the control treatment, which gave the lowest average yield of 21.090 g. The varieties also showed a significant effect on this trait, as the local variety was superior by giving the highest average yield of 29.950 g, with an increase rate of 26.20% compared to the Spanish variety, which gave the lowest average of 23.767 g, while the New Zealand variety recorded an average of 23.875 g. The binary interaction treatment between the local variety and 150 mg L^{-1} of copper oxide nanoparticles achieved the highest seed yield of 35.80 g, with an increase rate of 73.79% compared to the interaction between the Spanish variety and the zero concentration of copper oxide nanoparticles, which gave the lowest yield of 20.60 g. This physiological superiority is attributed to the vital role of copper nanoparticles in enhancing physiological processes associated with growth and production, especially improving photosynthetic efficiency and increasing the activity of enzymes responsible for the formation and translocation of nutrients from leaves to seeds, which led to increased dry matter accumulation, improved seed filling, and higher individual plant yield. Nano-copper also helped to increase nutrient absorption efficiency and improve hormonal balance within the plant, which was positively reflected on the number and weight of formed seeds. The superiority of the local variety is due to its possessing higher genetic capacity and distinctive physiological efficiency in utilizing nutrients and converting them into economic yield, as well as its better adaptation to local environmental conditions compared to the other varieties (3).

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Table (4). Effect of copper oxide nanoparticles on seed yield (g plant^{-1}).

N / V	N0	N1	N2	N3	Mean V
V1	22.00 g	29.62 c	32.40 b	35.80 a	29.950 a
V2	20.67 h	22.57 g	25.55 de	26.72 d	23.875 b
V3	20.60 h	23.25 fg	24.73 ef	26.48 d	23.767 b
Mean N	21.090 d	25.140 c	27.560 b	29.670 a	

Conclusions

This study provides decisive physiological and genetic evidence for the high efficiency of nano-fertilization technology using copper oxide in improving the productive and nutritional system of the faba bean plant. The study concluded that the use of copper oxide nanoparticles led to a clear improvement in the physiological and productive performance of the faba bean plant. Foliar spraying at a concentration of 150 mg L^{-1} contributed to raising the plant's content of essential nutrients (nitrogen, phosphorus, and potassium) and improving yield traits such as seed weight and individual plant yield, reflecting a stimulating role in enhancing the efficiency of metabolic processes, photosynthesis, and the translocation of assimilates toward the reproductive parts. The local variety was characterized by higher genetic efficiency in nutrient utilization and producing better yield compared to the New Zealand and Spanish varieties, while some traits such as the number of seeds per pod showed genetic stability and were not affected by fertilizer treatments. The results confirmed a clear synergistic effect when combining the local variety with the highest concentration of nano-fertilizer, indicating that the integration between genetic efficiency and nano-nutrition represents a critical factor in raising productivity and improving plant efficiency. Therefore, this technology can be adopted within modern sustainable agriculture strategies to improve faba bean production.

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