Research Article

Evaluation of the Effectiveness of Nano Zinc Oxide in Increasing Growth and Yield of Five Broad Bean (*Vicia faba* L.) Genotypes

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Abstract | In order to ascertain the degree to which nano-zinc oxide successfully improved the development and production of five broad bean genotypes, this study was carried-out in the winter of 2023–2024 in the research farm (Al-Mahawil area), Babil Governorate. Three replications of a randomized complete block design (RCBD) were used in a factorial experiment. Five broad bean genotypes sakiz bakr, luz de otno, dolce star, local, and faba daalta were the study's starting variables. The second element was nano-fertilization, which was administered directly to the plants at three distinct foliar application rates (0, 1, 2, and 3 mg L^{-1}). At the 5% probability level, means were compared using least significant difference testing. The following is a summary of the findings. According to statistical research, plants of the Dolce Star cultivar produced the highest mean values for the attributes of pod length, number of seeds in pods, and total seed production, reaching 21.967 cm, 8.733 (seeds.pods^{-1}), and 3.525 (tonnes H^{-1}) per plant, respectively. The Sakiz Bakr variety outperformed the others in terms of pod count, with the highest mean value of 12.650 (pods plant-1). However, with the greatest mean values for seed weight (100 seeds), seed protein content (157.167 g), and 30.500 (%), the variety Luz de Ottono performed better than the others. Plants fed with nano-fertilizer exhibited the highest percentage in all growth parameters at a dosage of 3 mg/l, according to results on pod length, number of pods, number of seeds per pod, weight of 100 seeds, total seed yield, and protein content in seeds. 149,000 (g), 3,553 (tonnes h^{-1}), 8,273 (pod seeds-1), 680 (pod plants-1), 22,253 cm, and 31,340 (%) are the pertinent results.

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Introduction

One of the primary winter legume crops, solanaceous beans (*Vicia faba* L.), are one of the most vital vegetable protein sources for human nutrition due to their high protein (25–40%) and high carbohydrate content. Thus, via nitrogen fixation processes in the soil, it not only significantly affects people's diets, particularly those with low incomes, but also enhances soil qualities (Ibrahim, 2011). Over the course of two seasons, Osman *et al.* (2013) assessed four distinct faba bean varieties. The findings indicated that although there were no significant variations in production or pod number (pods/plant-1), there were significant differences in seed quantity (seeds/plant-1) and 100-seed weight at the two sites. To increase manufacturing efficiency, a variety of techniques and tools were used. Among these techniques, one of the most crucial elements for raising yields is choosing the appropriate variety for the production region. This is true for many crop kinds in general. Mitiku and Wolde (2015) investigated eight bean cultivars in Ethiopia and discovered that they differed significantly in many characteristics, such as grass height, number of pods per plant, number of seeds per pod, 100-seed weight, unit yield per plant, total seed yield, and biological yield. Numerous researches have been conducted recently on the use of nanotechnology in agriculture. The research has concentrated on the application of nanotechnology to prevent plant diseases, preserve food, and cleanse soil of heavy metals (Awasthi *et al.*, 2017). These nanoparticles cause phenotypic and physiological changes in plants depending on their properties, chemical composition, size, surface coverage, concentration, and effectiveness depend on the plant species, such as its role in seed germination and plant growth (increasing plant biomass), the total number of vegetative branches, roots, and photosynthesis (Yahya, 2020). Zinc oxide nanoparticles (ZnO-NPs), one of the most common nanoparticles, have also been shown to improve growth and productivity in a number of leguminous plants (Yahya, 2019). They have also looked at how so-called nano-fertilizers may be used in lieu of traditional fertilizers to boost agricultural yields and encourage plant development (Veronica *et al.*, 2015). Because of its simple availability, low cost, durability at high temperatures, ability to absorb UV light, neutral pH, antimicrobial properties, and agricultural productivity, zinc oxide nanoparticles, or ZnO, have long been utilised extensively in a variety of sectors. They are gaining more attention in the agricultural sector because of their potential to boost growth and production (Drebee *et al.*, 2022). The number of pods, branches, blooming rate, and seed output all rose with zinc supplementation, according to (Jiang, 2014) (El-Fouly *et al.*, 2010). Applying a 2% zinc spray on broad beans enhanced seed quality and yield in addition to increasing the number of fruiting flowers. According to Thalooth *et al.* (2006), broad beans treated with the nutrient zinc sulphate produced more seeds, branches, and pods, decreased flower drop, and produced more fruiting flowers. One of the elements influencing crop development and establishment is the timing of fertilizer delivery; blooming and grain filling times have been shown to have a significant impact on pod production (Ali, 2011). The present study aimed to evaluate the growth and yield performance of five faba bean genotypes, determine the optimal concentration of nano zinc oxide for maximizing growth and productivity, and assess the interaction between genotype and nano-fertilizer treatments on plant performance.

Materials and Methods

During the winter of 2023–2024, the experiment was conducted in one of the farms in the (Babylon Governorate's Al-Mahawil district) to evaluate the effectiveness of nano zinc oxide in enhancing the growth and production of five distinct faba bean genotypes. Three randomized complete block design (RCBD) repetitions were used in the factorial experiment. The use of five faba bean genotypes (Local, Dolce star, Luz de otono, Faba Da orta, sakiz Bakl) is the first variation in the research. The second component of the investigation is the intermediate foliar spray application of nano-fertilization to the plants at three distinct doses (0, 1, 2, and 3 mg L^{-1}). The replicated number included 20 experimental units, with a total of 60 experimental units and an area of (2*3) m^2.

Zinc oxide (ZnO) nanoparticles >100 nm in size, weighing 81.39 g/ml, were obtained from Sigma-Aldrich, USA. Standard preparation was carried out by dissolving 0.25 g of zinc oxide powder in 250 ml of distilled water. The concentrations used in this study were (0, 1, 2, and 3 mg L^{-1}).

After preparing the concentrations used in the zinc spray solution, the vegetative system was sprayed in the early morning using a backpack sprayer. A spreading agent (liquid detergent) was added to the solution to reduce the surface tension of the spray solution and to ensure complete wetting of the plant at the stage of the beginning of flower bud emergence on February 6 for both seasons. The control treatment was sprayed with water only.

Table 1: *A physical and chemical characteristic of the experimental field's soil.*

Quantity mg kg^{-1} soil					EC m.dS^{-1}	pH	Organic matter g kg^{-1} soil	Soil Separators gm kg^{-1} soil			Soil texture
Fe	Zn	K	P	N				Sand	Clay	Loam	
3.8	0.40	177	10.4	64.2	3.96	7.5	11.11	160	280	560	Silty clay loam

There were three seeds in each hole, and after they had germinated, there were only two plants. Nitrogen was applied in two batches, one after germination and the other at the beginning of the blooming stage as urea fertilizer (46% N, 44 kg ha^{-1}) while phosphorus was supplied after planting (40 kg ha^{-1}) as triple superphosphate. Prior to planting, soil samples were taken from the top layer between 0 and 20 cm below the surface, and a number of physical and chemical properties were identified in the samples (Table 1). 135 days after planting, the number of field bears that destroyed the crop was counted:

1. Pod length (cm): A metre rule was used to measure the length of ten randomly chosen pods from each plot after they reached full maturity.
2. Number of pods in the plant (Pod plant-1): This characteristic was quantified by calculating the average number of pods per plant for each plot.
3. It was computed by dividing the number of seeds on the plant being observed by the number of pods of the same plant under the same circumstances; that is, the correct formula for calculating the number of seeds in a pod is: Number of seeds per pod = Total number of seeds per plant / Number of pods per plant.
4. Weight of 100 seeds: after combining the seeds that were taken from the plants and weighed on a sensitive scale, the estimated weight of 100 seeds (g) was determined from each treatment.
5. Total seed yield (tonne h^{-1}): Ten guarded plants were chosen at random from each testing unit's median lines. Based on plant density, the weight of the seeds was calculated and converted to tonnes h-1.
6. Seed protein content (%): The following formula indirectly calculates protein yield:

$$\textit{Total protein yield} = \textit{Seed yield tons } h^{-1} \times \textit{Percentage of protein in seeds}$$

Data were analyzed using GenStat software, and treatment means were compared using the Least Significant Difference (LSD) test at the 5% significance level ($p \leq 0.05$)

Results and Discussion

The Dolce star variety outperformed all genotypes tested, with the longest mean pod length of 21.967 cm, according to the results of the analysis of variance shown in Table 2. The Local variety, on the other hand, had the smallest mean pod length, measuring 14.683 cm. Furthermore, the control treatment produced the smallest mean pod length of 17.820 cm, whereas the treatment with nano fertilizer at 3 mg L^{-1} produced the most significant mean pod length of 22.253 cm, far surpassing the other fertilizer treatments. The interaction between variety and nano-fertilization was shown to have a significant effect on the pod length characteristic. In particular, the Dolce star coupling with 3 mg L^{-1} produced the greatest mean of 24.767 cm, which was much better than the other interactions, while the Local combination with 1 mg L^{-1} produced the lowest mean of 10.800 cm.

Table 2: *Genotypes and nanozinc's impact on pod length (cm).*

Mean	3	2	1	0	ZnO mg L^{-1} varity
14.683	16.133	16.133	10.800	14.100	Local
21.967	24.767	22.300	20.767	20.033	Dolce star
20.542	23.133	21.700	19.233	18.100	Luz de otono
21.383	24.400	22.200	19.700	19.233	Faba Da orta
19.458	21.267	20.167	18.767	17.633	sakiz Bakl
Interaction 2.932	22.253	20.500	17.853	17.820	Mean
	ZnO=1.388		Varity= 1.552		$L.S.D_{0.05}$

Plants of the Sakiz Bakl variety fared better than those of any other variety in the study, with an average of 12.650 pods (pod $plant^{-1}$). On the other hand, the Faba Da orta variety yielded the fewest pods, with a total of 8.808 (pod $plant^{-1}$). The research factors had a substantial impact, according to the statistical analysis shown in Table 3. While the nano fertilizer treatment at 3 mg L^{-1} produced the highest average of 12.680 (pod plant -1) among all fertilizer treatments, the control treatment produced the lowest average number of pods at 10.580 (pod plant -1). Pod production was impacted by the strong bidirectional relationship between fertilizer type and nano fertilizer; the interaction of Sakiz Bakl and 3 mg L-1 produced a significantly higher mean of 13.800 (pod plant -1), while the interaction of Luz de otono and the control produced the lowest mean of 8.100 (pod plant -1).

Table 3: *Impact of nano zinc and genotype on pod count.*

Mean	3	2	1	0	ZnO mg l^{-1}/ varity
11.600	12.800	12.100	11.400	10.100	Local
15.825	16.800	16.100	15.300	15.100	Dolce star
9.125	10.200	9.400	8.800	8.100	Luz de otono
8.808	9.800	9.200	8.433	7.800	Faba Da orta
12.650	13.800	12.800	12.200	11.800	sakiz Bakl
Interaction 0.182	12.680	11.920	11.227	10.580	Mean
	ZnO=0.082		Varity= 0.091		$L.S.D_{0.05}$

Table 4: *Impact of nano zinc and genotype on the quantity of seeds per pod.*

Mean	3	2	1	0	ZnO mg l^{-1} varity
6.067	7.133	6.400	5.833	4.900	Local
8.733	10.267	9.200	8.400	7.067	Dolce star
7.525	8.367	7.833	7.300	6.600	Luz de otono
7.367	8.067	7.767	7.167	6.467	Faba Da orta
6.525	7.533	6.933	6.233	5.400	Sakiz Bakl
Interaction 0.410	8.273	7.627	6.987	6.087	Mean
	ZnO= 0.183		Varity= 0.205		$L.S.D_{0.05}$

The Dolce star variety outperformed all the types tested, producing the greatest average number of seeds per pod at 8.733 (seeds per pod^{-1}), while the Local variety produced the fewest seeds per pod at 6.067. Table 4 analysis of variance results showed that the research factors had a substantial impact. With the greatest average of 8.273 seeds per pod (seeds per pod^{-1}), the treatment with nano fertilizer at 3 mg L^{-1} outperformed all other fertilizer treatments, according to the results, while the control treatment had the lowest average of 6.087 seeds per pod (seeds per pod^{-1}). The quantity of seeds per pod was dramatically impacted by a noticeable two-way interaction between the cultivar and the nano fertilizer. The greatest mean, 9.200 (pod seeds-1), was obtained when Dolce star and 2 mg L^{-1} were combined, while the lowest mean, 4.900 (pod seeds-1), was obtained when Local and control were combined.

The local cultivar had the lowest mean weight of 100 seeds at 108.500 g, while the cultivar Luz de otono showed the best performance in the research with the highest mean weight of 100 seeds at 157.167 g. The study factors had a substantial impact on the outcomes, according to the analysis of variance shown in Table 5. With a mean weight of 149.000 g, the treatment that used nano fertilizer at a concentration of 3 mg L^{-1} generated the greatest mean weight of 100 seeds, whereas the control treatment produced the lowest mean weight of 100 seeds, 122.933 g. Additionally, the weight of 100 seeds was impacted by a significant two-way interaction between the cultivar and the application of nano-fertilization; the combination of Luz de otono and 3 mg L^{-1} produced the highest mean of 168.000 g, while the combination of Local and control produced the lowest mean of 95.000 g.

Table 5: *Impact of nano zinc and genotype on 100-seed weight.*

Mean	3	2	1	0	ZnO mg l^{-1} / varity
108.500	124.333	114.667	100.000	95.000	Local
128.500	145.333	134.333	122.667	111.667	Dolce star
157.167	168.000	161.667	155.000	144.000	Luz de otono
153.500	163.333	158.333	149.667	142.667	Faba Da orta
133.250	144.000	137.333	130.333	121.333	sakiz Bakl
Interaction 5.453	149.000	141.267	131.533	122.933	Mean
	ZnO=2. 644		Varity=2.956		$L.S.D_{0.05}$

Table 6: *Total seed yield (tones ha^{-1}) as a function of genotype and nano zinc.*

Mean	3	2	1	0	ZnO mg l^{-1}/ varity
2.525	3.133	2.800	2.300	1.867	Local
2.842	3.500	3.267	2.300	2.300	Dolce star
3.525	3.867	4.000	3.233	3.000	Luz de otono
3.208	3.800	3.533	2.800	2.700	Faba Da orta
2.767	3.467	2.833	2.500	2.267	sakiz Bakl
Interaction 0.343	3.553	3.287	2.627	2.427	Mean
	ZnO=0.155		Varity= 0.173		$L.S.D_{0.05}$

With the greatest average for seed yield characteristics (3.525 tonnes per hectare), the Dolce Star variety of plants did better than any other variety in the trial. On the other hand, with just 2.525 tonnes per hectare, the local variety yielded the least amount of seed overall. The research factors had a substantial impact, according to the analysis of variance data shown in Table 6. Additionally, outperforming all other fertilizer treatments, the nano fertilizer treatment at 3 mg L^{-1} produced the greatest mean total seed yield of 3.553 tonnes per hectare, whereas the control treatment produced the lowest mean total seed yield of 2.427 tonnes per hectare. Furthermore, total seed output (tonne h^{-1}) was considerably impacted by the variety-nano-fertilization relationship. The combination of Luz de Otono with 2 mg L^{-1} produced the greatest mean at 4.000 (tonne h^{-1}), while the interaction of Local with control produced the lowest mean of 1.867 (tonne h^{-1}).

Table 7: *Genotype and nanozinc's effects on seed protein content (%).*

Mean	3	2	1	0	ZnO mg l^{-1}/ varity
23.417	30.000	23.667	21.333	18.667	Local
27.917	32.667	30.333	27.333	21.333	Dolce star
30.500	34.667	32.333	29.000	26.000	Luz de otono
27.675	30.700	29.333	26.333	24.333	Faba Da orta
25.833	28.667	27.333	24.333	23.000	Sakiz Bakl
Interaction 3.211	31.340	28.600	25.667	22.667	Mean
	ZnO=1.944		Varity=1.173		$L.S.D_{0.05}$

The findings reported in Table 7 from the analysis of variance reveal that the cultivar Luz de Otono surpassed all other studied cultivars, obtaining the greatest mean seed protein content at 30.500 (%). In comparison, the Local cultivar produced a lower seed protein content of 23.417 (%). Regarding nano-fertilizer application, the treatment with 3 mg L^{-1} of nano-fertilizer greatly topped all other fertilizer treatments, resulting in the highest seed protein content of 31.340 (%). Conversely, the control treatment had the lowest mean seed protein content at 22.667 (%). The interaction between the variety and nano-fertilization considerably altered the seed protein content, as indicated by the Luz de Otono + 3 mg L^{-1} combination, which reached a remarkable mean of 34.667 (%), much above the other relationships. In contrast, the Local + control interaction resulted in the lowest mean of 22.667 (%).

The results of this study showed significant differences among the faba bean genotypes in all measured characteristics. Plants of the cultivar Dolce star scored better in delivering the highest mean among criteria of pod length, number of seeds in pods, and total seed production. Tables 2, 3, 6. The different natures of the genes of the two varieties, and the differences in the genetic factors of the two varieties might be the reason behind the dibs on fava bean trait because most of the studies indicate that there is a significant variation among fava bean varieties for seeds/pod trait and it is more due to genetic factors than the concomitant growth factors; moreover, different varieties might take different considerations of these traits. Differential varieties in genetic characters or varieties in growth habit contribute to expressivity in the number of seeds per pod; either number might also be due to the availability of nutrients for the seeds and lack of competition among them, which indicates that where there is no individual competition for pod and each seed, the chances of seed abortion and incomplete fertilization are rather reduced which ultimately leads to more seeds and therefore a higher total seed yield. The Sakiz Bakl variety plants have increased genetic diversity amongst kinds in terms of the quantity of pods in the plant (Ayed, 2012) acquired large variance mean of seeds among types of broad bean crop pods, it can be claimed that the likely influence of zinc is reflected in lowering female gametophyte abortion rates, therefore seed numbers observed. Hence, this will lower seed abortion occurrences and enhance the metabolism of proteins, carbohydrates, and specific growth regulators, awakening seed number of sprayed bean plants in a single pod. These results were in accordance with (Al-Shamma, 2014; Negash *et al*., 2015; Al-Myali *et al*., 2020; Hussain *et al*., 2019; Alnuaimi *et al*., 2019). Where the Luz de Ottono plants succeeded to offer the greatest mean in the 100-seed weight characteristic and seed protein content, seed weight of other bean types differed. The mean 100-seed weight advantage of this feature might be expressed from the variations in seed size across genotypes. Any plant must completely inflate its seeds, which relies on the available photosynthetic rate and the translocation of its byproducts. In superiority, this may be attributed to the plant reaching its maximum development, good vegetative growth getting induced, nutrients being produced in the optimum quantities (Hassan *et al*., 2021), and their storage and transportation to the estuary during seed filling, as well as the related physiological and life processes that bear upon the capability of exploiting the products of carbon synthesis and add to the buildup of these components in the seeds. This reflected heightened weight and protein content of seeds (Fageria, 2009; Ibrahim, 2011b; Al-Musawi, 2013) suggested a rise, which by all measures was a favourable indicator of this. Pod length, pod number,

number of seeds per pod, weight of 100 seeds, total seed output, and protein content in seeds were greatest in nano fertilizer 3 mg L^{-1} treated plants. Tables 2, 3, 4, 5, 6, 7 Zinc plays a vital role in auxin biosynthesis, enzyme activation, protein metabolism and membrane stability. Zinc is engaged in several vital processes and activates numerous enzymes. Besides, it is crucial in the synthesis of auxin among other reasons why the plant wide beans of O'neill *et al.* (2004) recorded longer pods. This tendency correlates with the results of Abd El-azeem *et al.* (2012) that imply that increase in the number of pods will lengthen the life of the chlorophyll pigments via a lower oxidation state (Abu El-Yazied *et al.*, 2012; Al-Zubaidi, 2024). Zinc also promotes the synthesis of carbs. A. R. and how it develops into the age of accelerating photosynthesis (Jiang *et al.*, 2014) and from zinc's participation in synthesizing membranes in cells and protecting them from being oxidized due to definite types of oxygen reactions, which affect the flow and movement of different materials within the roots, and eventually to the entire plant (Myali *et al.*, 2000) for each kind and degree of its reactivity to That fecundity was ascribed to more blooms being set and, consequently, resulting to more pods. The findings of Ibrahim (2011b) and Issa *et al.* (2019) findings, which indicated the number of pods on a bean plant significantly increase upon zinc and boron application provides evidence that indeed the increased number of pods by the plant come up due to increased concentration of the elements on the plant. Outcome and grain yield expression of zinc may be linked to in promoting and speeding photosynthesis meant that proteins and carbohydrates would be created, thus boosting the accumulation of dry matter. It is also important for pollen productivity and tube activity thus assuring pods would grow packed with the most developed and ripe seeds (Rizk and Abdo, 2001). The zinc spray treatments affected Attending resulted in diminished competition between seeds for the food manufactured inside the plant therefore more flowers were fertilized wherein seed abortion might increase due to the increasing flowers that are pollinated and in play, and decreased competition between pods, as well-formed harboring increased nutrient concentration and filling the single pod. This was demonstrated by earlier research where they revealed A direct association between increasing the content of zinc with the weight of the seeds within the pod (Al-Isawi and Khrbeet, 2011; Myali *et al.*, 2020; Ali *et al.*, 2021). The enhanced sprouting dimensions of vegetative growth, emission of pod-bearing branches, and escalation of the characteristics of each individual plant resulting from zinc spraying contribute to the improvement in production per individual plant. One of the explanations would presumably be that the increase in seed yield was well mirrored by the rise in its component components accessible (Tables 2, 3, and 4). These findings are confirmative. This should be acknowledged as a consequence of the increase of the pods, plants, seeds, and other agricultural components with zinc. Because the seeds aspire to greater plant yields and better seed protein content, it is pretty apparent from the Pod Tables 3, 4 that the increment in the component components of the crop would ultimately improve the overall seed yield per unit area (El-Gizawy and Mehasen, 2009; Hassoon *et al.*, 2023).

Conclusions and Recommendations

Based on the results, we may make the following deductions:
Comparing to other kinds, the variety that produces the greatest mean total seed output is more likely to be grown than the plants of the variety (Dolce star). Pod length, number of pods, number of seeds per pod, weight of 100 seeds, total seed production, and percentage of protein in seed reached maximum values with plants supplied nanofertilizer at 3 mg L^{-1}. Cultivars with interaction of the nano-fertilizer (Luz de Otoño + 2 mg L^{-1}) performed better than others and had the highest means for the characteristic of total seed yield.

Novelty Statement

This study shows for the first time that the interaction between the 'Luz de Otoño' variety and 2 mg. L^{-1} nano-fertilizer significantly enhances seed yield compared to the rest of the treatments.

Author's Contribution

Ali S. Hassoon: Designed the experiments, supervised fieldwork and contributed to data collection.
Ahmad Thamer Kamil: Conducted data analysis, interpreted the results and wrote the initial draft.
Rafal Salih Mahdi: Assisted in lab work, reviewed literature and edited the final manuscript.

Conflict of interest
The authors have declared no conflict of interest.

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