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EFFECT OF MICRONUTRIENTS AND NANO FERTILIZERS ON THE GROWTH AND PRODUCTIVITY OF ROSELLE (*HIBISCUS SABDARIFFA* L.)

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SUMMARY

The presented study sought to investigate the response of roselle (*Hibiscus sabdariffa* L.) plants on the influence of micronutrients nano fertilizer (silicon and zinc), carried out in June 2021 on private orchard fields in the Wasit Province, Iraq. The treatment construction was a perfectly randomized factorial experiment ($5 \times 5 \times 3$) in a completely randomized design, done with three replications. The roselle plant received micronutrients spraying with nano fertilizer silicon at concentrations of 0, 3, 6, 9, and 12 ppm and nano zinc with concentrations of 0, 500, 1000, 1500, and 2000 ppm, studying their effects on the plant height (cm), fresh and dry weights (g plant^{-1}), anthocyanin content, total soluble sugars, acidity citric acid, and TFAA of roselle rhizome in yield in single and two-way interactions. The discovery revealed a single application of the previously indicated factors had a considerable influence on the roselle's development and productivity features, especially at high concentrations. Different outcomes resulted using two-way interactions. The most crucial effect observed was on the total soluble sugars. Based on the results, adding nano silicon and nano zinc to the roselle plant significantly increased all traits, regardless of the vegetable characteristics or chemical content achieved with the highest concentrations using the highest effects, whether alone or with two interactions.

Keywords: Roselle, micronutrients, nano fertilizer, silicon, zinc, total free amino acids

Key findings: In the roselle plants, adding nano silicon and nano zinc and their interactions highly improved all the vegetative and biochemical traits.

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INTRODUCTION

Roselle (*Hibiscus sabdariffa* L.) plants are part of the family Malvaceae, with primary cultivation for the plant's valuable economic component, the fleshy sepals (calyx) that surround the fruit (capsules) (Amin and Kanimarani, 2020). With a deep penetrating taproot, variegated green-to-red leaves, and large, short-peduncled, red-to-yellow flowers featuring a dark center, this annual summer shrub is erect and primarily branched. Effective cultivation of roselle occurs in the tropics and subtropics. The immature leaves serve as a green vegetable for consumption, and the primary product, the calyx, finds application in beverages. As a result, roselle has wide use in the culinary, cosmetic, and pharmaceutical industries, with its fruits and flowers helping heal bronchitis and coughing. The calyces also contain sugars, minerals, vitamin C, carotene, anthocyanins, flavonoids, steroids, triterpenoids, alkaloids, and valuable micronutrients. Moreover, they contain calories, protein, fiber, and fixed lipids, which account for the remaining 17% (Abou-Sreea *et al.*, 2022). Studies of roselle have long existed, including comparing foliar spraying with micronutrient nano fertilizer (silicon and zinc) on growth and productive characteristics.

In recent years, effective nano fertilizers or coated nano-nutrients have emerged to accelerate crop growth and nutrient release on demand, control nutrient release that regulates plant development, and increase target function. Nano-materials are materials with particle sizes ranging between 1 and 100 nm in at least one dimension. Thus, NMs may supply one or more nutrients to promote plant development while using a less traditional fertilizer and releasing nutrients slowly according to the crop growth curve. Nano fertilizers can enable quick plant response, especially when dealing with soil issues, such as excessive pH, carbonate minerals, and inadequate root development. Some research has shown that active nano fertilizers have advantages in terms of greater nutrient efficiency, higher yield, better quality, and a safer environment (Al-juthery and Saadoun, 2018).

Nano fertilizers may be the ideal solution for easing macro- and micronutrient deficits by improving nutrient absorption quality and dealing with chronic eutrophication problems. By employing nanomaterials covered with a thin coating or supplied as emulsions, nutrients can have better absorption in nano fertilization. The slow release of nutrients from nanoparticle-coated fertilizers enhances crops' nutrient uptake (Aljanabi, 2021). Nano-silicon particles (Si-NPs) have different physical and chemical characteristics than bulk materials and function better in reducing various biotic stresses than bulk materials (Abdel-Haliem *et al.*, 2017). The Si-NPs have tremendous potential in agriculture, and foliar application with silicon reduces the damaging effects of drought conditions, as it plays an essential part in reducing water stress by decreasing the rate of transpiration, enhancing the photosynthesis process by improving light receiving efficiency, keeping the leaf rigid and erect (Dawa *et al.*, 2020). Given the importance of Zn as a micronutrient for plants and human development, zinc oxide (ZnO) is an example of NP that might be advantageous in this respect. Plant development mainly depends on Zn nutrition, while Zn is a structural component of several proteins and enzymes. As they are more efficient and cost-effective than synthetic Zn fertilizers, zinc NFs of the ZnO type have more utilization in current agriculture (Seleiman *et al.*, 2020). However, trace minerals, such as Zn, may delay plant development when provided at high concentrations by causing specific metabolic changes in plants. Studies have found Zn NFs promote germination, seedling development, yield, and crop quality; however, they may also adversely affect growth parameters. In summary, Zn NFs stimulate growth, enhance production and efficiency, and are less expensive than synthetic fertilizers (Meen and Kumar, 2017).

MATERIALS AND METHODS

Experimentation commenced in June 2021 at the private orchard field in Wasit Province, Iraq. The seeds, originating from the Sinak

region, Baghdad, Iraq, underwent cultivation in compartmentalized containers filled with peat moss (Van Egmond), with a high concentration of vital components, a pH of 6, and an electrical conductivity between 1.6 and 1.8. Dividing compartments into three blocks, each further divided into 15 panels. There were 75 panels in all, with 10 seeds in each. Spraying of micronutrients nano fertilizer silicon at concentrations of 0, 3, 6, 9, and 12 ppm and nano zinc at concentrations of 0, 500, 1000, 1500, and 2000 ppm transpired on the roselle plant. Foliar spraying at one time ensued for the treatments in the vegetative stage of the plant.

After 21 days of foliar treatments, the study calculated the average plant height, fresh and dry weights (g plant^{-1}), and anthocyanin (Abdel-Aal and Hucl, 1999). Total soluble sugars determination by the phenol-sulfuric acid followed the method described by Saad *et al.* (2021). Titratable acidity assessment occurred for the percentage of citric acid present. The measurement procedures followed the guidelines of the Association of Official Analytical Chemists (AOAC, 2002). Determining total free amino acids (TFAA) concentration (mg g^{-1} DW) colorimetrically was according to Jayarman (1981).

Statistical analysis

The randomized complete block design (RCBD) analysis of variance test helped examine how statistically different treatments differed. The least significant difference (LSD) at 0.05% levels assessed mean comparison (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Plant height

Table 1 shows that nano silicon considerably influenced average plant height. The mean height of the plants increased steadily from 227.40 to 239.80 cm when the nano silicon concentration increased from 3 ppm to 12 ppm. Plants treated with nine and 12-ppm

nano silicon grew the highest. The nano zinc also significantly affected the average plant height. Similarly, when boosting the concentration of nano zinc, the average plant height grew progressively (Aipeisova *et al.*, 2023; Khan *et al.*, 2023). Compared with 500 ppm (227.67 cm) and the control (222.93 cm), nano zinc at 2000 ppm generated massive plants (239.13 cm). The interaction effect between micronutrients of nano fertilizer (silicon and zinc) showed significant (Table 1). The tallest plants emerged from being sprayed with 12 ppm of nano silicon and 2000 ppm of nano zinc.

Plant fresh weight

Table 2 shows that nano silicon substantially affected the plant's average fresh weight. The mean fresh weight of the plants increased steadily from $2.3733 \text{ g plant}^{-1}$ to $2.4520 \text{ g plant}^{-1}$ when the nano silicon concentration increased from 3 to 12 ppm. The plants with the highest fresh weight were those treated with 12 ppm nano silicon. Likewise, the nano zinc significantly influenced the average fresh weight of the plant (Goswami *et al.*, 2022). Similarly, when the concentration of nano zinc grew, the average fresh weight of plants climbed progressively. The highest concentration of nano zinc generated immense plants ($2.4440 \text{ g plant}^{-1}$) compared with 500 ppm ($2.3667 \text{ g plant}^{-1}$) and the control ($2.3073 \text{ g plant}^{-1}$). The interaction impact of micronutrients nano fertilizer (silicon and zinc) on the average fresh weight of the plant appeared significant (Table 2). Plants with a fresh weight of $2.4933 \text{ g plant}^{-1}$ resulted from treatments with 12 ppm nano silicon and 2000 ppm nano zinc.

Plant dry weight

The average dry weight of the plant gained significant impacts from nano silicon, as indicated in Table 3. The mean dry weight of the plants showed a consistent increase from $570.83 \text{ g plant}^{-1}$ to $633.05 \text{ g plant}^{-1}$, caused by the rise in nano silicon concentration from 3 to 12 ppm. The plants that received a 12 ppm nano-silicon spray had the maximum dry

Table 1. Effect of foliar application of nano silicon and nano zinc fertilization and their interactions on plant height.

Nano silicon	Nano zinc					Means (cm)
	0	500	1000	1500	2000	
0	214.00	215.67	221.00	226.00	226.67	220.67
3	218.33	223.33	231.67	229.67	234.00	227.40
6	222.33	227.33	234.67	235.67	240.33	232.07
9	229.33	236.00	242.67	244.00	247.00	239.80
12	230.67	236.00	241.00	243.67	247.67	239.80
Means (cm)	222.93	227.67	234.20	235.80	239.13	

LSD_{0.05} Nano zinc = 1.378, Nano silicon = 1.378, Interaction = 3.081

Table 2. Effect of foliar application of nano silicon and nano zinc fertilization and their interactions on the fresh weight of the plant.

Nano silicon	Nano zinc					Means (g plant ⁻¹)
	0	500	1000	1500	2000	
0	2.1033	2.2933	2.3300	2.3633	2.4167	2.3013
3	2.3067	2.3533	2.3933	2.4067	2.4067	2.3733
6	2.3367	2.3800	2.3833	2.4233	2.4267	2.3900
9	2.3667	2.3900	2.4300	2.4533	2.4767	2.4233
12	2.4233	2.4167	2.4567	2.4700	2.4933	2.4520
Means (g plant ⁻¹)	2.3073	2.3667	2.3987	2.4233	2.4440	

LSD_{0.05} Nano zinc = 0.03643, Nano silicon = 0.03643, Interaction = 0.08146

Table 3. Effect of foliar application of nano silicon and nano zinc fertilization and their interactions on the dry weight of the plant.

Nano silicon	Nano zinc					Means (g plant ⁻¹)
	0	500	1000	1500	2000	
0	522.95	532.25	547.28	555.50	567.09	545.01
3	545.15	565.15	572.50	583.95	587.39	570.83
6	563.06	576.86	581.71	594.23	598.88	582.95
9	555.50	583.50	602.50	621.29	631.31	598.82
12	574.09	588.69	650.12	670.21	682.16	633.05
Means (g plant ⁻¹)	552.15	569.29	590.82	605.03	613.36	

LSD_{0.05} Nano zinc = 3.842, Nano silicon = 3.842, Interaction = 8.590

weight. The typical dry weight of plants received considerable impacts from the nano zinc. Similarly, when the concentration of nano zinc climbed, the average dry weight of the plant steadily increased. Compared with 500 ppm (569.29 g plant⁻¹) and the control (552.15 g plant⁻¹), 2000 ppm of nano zinc produced the highest plant weight (613.36 g plant⁻¹). The interaction impact between micronutrients nano fertilizer (silicon and zinc) occurred significantly on the average dry weight of the plant (Table 3). The dry weight of plants was 682.16 g plant⁻¹ treated with 12 ppm of nano

silicon and 2000 ppm of nano zinc (Suciatty *et al.*, 2018).

Anthocyanin content

The nano silicon significantly impacted the average anthocyanin concentration of leaves, as shown in Table 4. The mean anthocyanin content on plant leaves showed an upward trend from 25.750 to 30.075 mg g⁻¹ DW due to the increase in nano silicon concentration from 3 to 12 ppm. Plants treated with 12 ppm of nano silicon had the leaf's highest anthocyanin

content (Abou-Sreya *et al.*, 2022). The nano zinc also affected significantly the average anthocyanin concentration of leaves. Similarly, when the concentration of micro zinc ascended, the average anthocyanin content on leaves steadily increased. Compared with 500 ppm, 2000 ppm of nano zinc generated anthocyanin content on leaves of plants at 29.877 mg g⁻¹ DW, as opposed to 25.765 mg g⁻¹ DW. The interaction impact between micronutrients nano fertilizer (silicon and zinc) proved substantial on the average anthocyanin concentration of leaves (Table 4). The leaves sprayed with 12 ppm of nano silicon and 2000 ppm of nano zinc had the highest anthocyanin content (35.207 mg g⁻¹ DW).

Soluble sugars

Table 5 indicates that nano silicon significantly affected average total soluble sugars. The mean total soluble sugars of the plants showed a consistent increase from 0.4467% to 0.4467% due to the rise in nano silicon concentration from 3 to 12 ppm. Plants treated with 12 ppm of nano silicon had the maximum

total soluble sugar levels. The average total soluble sugars received significant impacts from the nano zinc. The average total soluble sugars steadily increased as the nano zinc concentration increased. Formation of total soluble sugars has a rate of 0.4953% at 2000 ppm of nano zinc, compared with 0.4427% and 0.4167% at 500 ppm and the control, respectively. The interaction impact between micronutrients nano fertilizer (silicon and zinc) emerged substantial on the average total soluble sugars (see Table 5). The total soluble sugars included those exposed to nano silicon and nano zinc at 12 ppm and 2000 ppm, respectively.

Acidity of citric acid

Table 6 shows the nano silicon significantly affected the average acidity of citric acid. The mean acidity of the plants' citric acid steadily increased from 0.4893% to 0.5633% due to an increase in nano silicon concentration from 3 to 12 ppm. Plants treated with 12 ppm of nano silicon produced the highest citric acid acidity.

Table 4. Effect of foliar application of nano silicon and nano zinc fertilization and their interactions on the anthocyanin content of leaves.

Nano silicon	Nano zinc					Means (mg g ⁻¹ DW)
	0	500	1000	1500	2000	
0	22.467	22.913	23.053	24.467	25.320	23.644
3	23.813	25.227	25.773	26.747	27.190	25.750
6	24.153	26.227	27.553	28.850	29.317	27.220
9	25.413	27.177	30.133	30.577	32.353	29.131
12	25.873	27.280	29.910	32.103	35.207	30.075
Means (mg g ⁻¹ DW)	24.344	25.765	27.285	28.549	29.877	

LSD_{0.05} Nano zinc = 0.4644, Nano silicon = 0.4644, Interaction = 1.038

Table 5. Effect of foliar application of nano silicon and nano zinc fertilization and their interactions on total soluble sugars.

Nano silicon	Nano zinc					Means (%)
	0	500	1000	1500	2000	
0	0.3967	0.4033	0.4133	0.4233	0.4433	0.4160
3	0.3967	0.4300	0.4500	0.4733	0.4833	0.4467
6	0.3967	0.4300	0.4567	0.4767	0.4800	0.4480
9	0.4433	0.4800	0.4900	0.5067	0.5200	0.4880
12	0.4500	0.4700	0.5200	0.5200	0.5500	0.5020
Means (%)	0.4167	0.4427	0.4660	0.4800	0.4953	

LSD_{0.05} Nano zinc = 0.00893, Nano silicon = 0.00893, Interaction = 0.01997

Table 6. Effect of foliar application of nano silicon and nano zinc fertilization and their interactions on the acidity of citric acid.

Nano silicon	Nano zinc					Means (%)
	0	500	1000	1500	2000	
0	0.4167	0.4267	0.4500	0.4633	0.4833	0.4480
3	0.4567	0.4733	0.4800	0.5067	0.5300	0.4893
6	0.4600	0.4700	0.5333	0.5500	0.5600	0.5147
9	0.4633	0.5000	0.5533	0.5733	0.6067	0.5393
12	0.5033	0.5400	0.5600	0.5867	0.6267	0.5633
Means (%)	0.4600	0.4820	0.5153	0.5360	0.5613	

LSD_{0.05} Nano zinc = 0.00893, Nano silicon = 0.00833, Interaction = 0.01863

Table 7. Effect of foliar application of nano silicon and nano zinc fertilization and their interactions on total free amino acids (TFAA).

Nano silicon	Nano zinc					Means (mg g ⁻¹ DW)
	0	500	1000	1500	2000	
0	102.59	108.39	110.12	113.32	115.22	109.93
3	116.10	120.94	124.68	125.89	128.38	123.20
6	118.00	121.59	129.09	131.34	133.69	126.74
9	115.22	123.07	131.68	133.36	136.13	127.89
12	120.02	131.25	130.19	138.65	138.74	131.77
Means (mg g ⁻¹ DW)	114.39	121.05	125.15	128.51	130.43	

LSD_{0.05} Nano zinc = 1.369, Nano silicon = 1.369, Interaction = 3.061

The average citric acid acidity also attained significant impacts from the nano zinc. As the quantity of micro zinc grew, the average acidity of citric acid steadily increased. The citric acid generated at 2000 ppm of nano zinc had an acidity of 0.5613% compared with 500 ppm's 0.4820% and the control's 0.4600%. The interaction impact between micronutrients nano fertilizer (silicon and zinc) and the average citric acid emerged to be substantial (see Table 6). The citric acid sprayed with 12 ppm of nano silicon and 2000 ppm of nano zinc had the highest acidity (0.6667%).

Free amino acids

Table 7 suggests that microsilicon significantly affected the citric acid's average acidity. The mean acidity of the citric acid in plants showed a consistent rise from 123.20 to 131.77 mg g⁻¹ DW because of the increase in nano silicon concentration from 3 to 12 ppm. The citric acid with the utmost acidity was evident in plants treated with 12 ppm of nano silicon. The nano zinc also influenced significantly citric acid's average acidity. Likewise, as the quantity of

micro zinc grew, the average acidity of citric acid steadily increased. The citric acid generated at 2000 ppm of nano zinc was 130.43 mg g⁻¹ DW in acidity compared with 121.05 mg g⁻¹ DW and 114.39 mg g⁻¹ DW at 500 ppm and the control, respectively. On the average acidity of citric acid, the interaction effect between micronutrients nano fertilizer (silicon and zinc) revealed significant (Table 7). The acidic citric acid incurred spraying with 12 ppm of nano silicon and 2000 ppm of nano zinc (138.74 mg g⁻¹ DW).

The effects of micronutrient nano fertilizer (silicon and zinc) on plant physiological characteristics provided positive impacts (Tables 1, 2, and 3). The nano fertilizers are nutrient carriers of NPs ranging in size from 30–40 nm, capable of keeping nutrient ions due to specific properties, such as high surface area, high absorption quality, increased 20%–200% production rate, increase in chlorophyll content, and expansion of leaf surface area (Goswami *et al.*, 2022). Silica's shape of the surface enables it to function as a nutrient carrier, with the potential to transform agriculture's nutrient delivery

system. Nanosilica's application, apart from the fact that mesoporous produced substantially higher results regarding nutritional fortification, enhanced production, and nutrient utilization efficiency (Durgude *et al.*, 2022). Silicon helps maintain mineral balance, simultaneously enhancing root water absorption, limiting harmful ion uptake, and ensuring the effective functioning of anti-oxidative mechanisms (Dawa *et al.*, 2020).

The addition of ZnO-NPs caused an increase in seedling length. Nanoparticles have the potential to enhance seedling germination and development by stimulating enzymatic activity involved in various metabolic pathways and biochemical functions. Similarly, nanotechnology treatments have proven to improve crop health and yield sustainably (Yadav *et al.*, 2023). The effects of micronutrient nano fertilizer (silicon and zinc) on plant vegetative characteristics sustained positive influences (Tables 4, 5, 6, and 7). In addition, SiO₂ NPs increase photosynthetic pigments by increasing endogenous cytokinin levels, which improve the chloroplast ultrastructure. In addition, increasing carbohydrate content may enhance osmotic pressure in plants, allowing for better absorption of soil water and spraying with NP-containing solutions, increasing osmolyte levels. The SiO₂ is also essential in balancing element intake, transport, and distribution through water uptake and root growth (Jasem and Khalil, 2022).

Silicon has increased plant growth, mineral nutrition, mechanical strength, and fungal disease resistance. The nanoparticles are often created as suspensions and sprayed onto plants or by soaking the plants in suspension to determine the influence of nanoparticles on plants, affecting the plant's development and productivity, particularly the active substances (Li *et al.*, 2023). Nano-silica fertilizer effectively influences the leaves, causing them to grow erect and extend deep into the leaf surface, receiving more sunlight and optimal photosynthesis. Therefore, producing more photosynthate stored in the plant's vegetative organs, such as roots, stems, and leaves, as a food reserve for plant development (Suciety *et al.*, 2018).

CONCLUSIONS

According to the results, adding nano silicon and nano zinc to the roselle plant significantly increased all traits, regardless of whether the characteristics of vegetable or chemical content achieved the highest concentrations used the highest effects, whether individually or as two interactions.

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