

Physiological and Biochemical Processes in the Exogenous Administration of Selenium Nanoparticles and Selenium/Copper Oxide Nanocomposite to Reduce Salt Stress in *Zea mays* L.

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Research on nanoparticles (NPs) is gaining increasing popularity as a way to enhance abiotic stress tolerance and improve crop productivity. This study assessed the effects of foliar spray of selenium NPs (Se NPs) and selenium/copper oxide nanoparticles (Se/CuO NPs) at 50 and 100 ppm on the growth and biochemical characteristics of *Zea mays* L. plants grown under saline stress conditions (100 mM). Se NPs and Se/CuO NPs were analyzed by energy dispersive X-ray, transmission electron microscopy, and Fourier transform infrared spectroscopy analyses. The Se NPs and Se/CuO NPs were found to have an average particle size of 135.2 and 75.1 nm using the ImageJ tool. Shoot and root lengths, chlorophyll levels, protein, phenols, and flavonoids were all investigated in this study. Plant growth and chlorophyll concentration dropped under salt stress but were improved with the application of Se and Se/CuO NPs. The enzymes catalase, superoxide dismutase, and glutathione reductase exhibited the highest values at 100 ppm of Se/CuO NPs, of 74.4, 132.1, and 43.2 mmol/g, respectively. Se and Se/CuO NPs reduced stress and increased chlorophyll. ZnO-NPs improved maize plants' resistance to the unfavorable effects of saline soils. Finally, plant metabolism and abiotic stress tolerance were improved by Se and Se/CuO NPs.

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INTRODUCTION

Salinity stress is a significant issue in crop production and a crucial limiting factor on a global scale. In dry and semi-arid regions of the world, salinity is the most prevalent abiotic stressor (Hussain *et al.* 2019). Plants' morpho-physiological characteristics are negatively impacted by salinity, which lowers their production. Osmotic stress brought on by salinity reduces plant growth and productivity in irrigated areas. Plants under salinity stress experience changes in their ability to absorb water and disturbances in their ionic composition (Ayub *et al.* 2021). An overabundance of salt in the soil causes early leaf senescence by decreasing the area of the leaf and the amount of chlorophyll, which lowers photosynthetic activity (Nawaz *et al.* 2019).

Following rice and wheat, maize (*Zea mays* L.) is the third most significant cereal crop in the world. It is cultivated worldwide and exhibits higher genetic diversity among

cereals due to its strong nutritional value. Numerous abiotic stressors negatively impact maize crop productivity (Revilla *et al.* 2022). One of the eight necessary micronutrients, copper (Cu) plays a crucial role in the formation of metalloproteins by regulating enzyme activity as a cofactor. It plays a key function in controlling the combination of several macromolecules that are essential to plant metabolism, such as respiration, photosynthesis, and various defensive mechanisms against abiotic stressors (Lodde *et al.* 2021). Chlorophyll concentration and seed dormancy are two aspects of plant growth and development that copper is crucial for regulating (Wang *et al.* 2021). Nanomaterials have recently gained traction across various fields as a solution to numerous environmental, medical, and agricultural issues (Alsalamah *et al.* 2023b; Qanash *et al.* 2023; Alghonaim *et al.* 2024; Amin *et al.* 2025).

Several studies have demonstrated that nanoparticles (NPs) may exert toxic and stress-inducing effects on biological systems (Wu *et al.* 2023; Yadav *et al.* 2023). The extent of these toxicological impacts depends on multiple parameters, including particle size, methods of synthesis, concentration, surface charge, and exposure time (Abdelghany *et al.* 2018). Despite their diverse and beneficial biological functions, it is essential to establish a balance between their positive and adverse effects to ensure safe utilization. Consequently, refining synthesis techniques and employing appropriate surface modifications are key approaches to mitigate NP toxicity (Sanità *et al.* 2020). Notably, low concentrations of selenium (Se) and copper oxide (CuO) nanoparticles have been reported to promote plant growth and alleviate stress responses (Hernández-Hernández *et al.* 2019).

Microorganism and plant extract synthesis has drawn a lot of interest as a sustainable, environmentally friendly method of producing a variety of NPs. (Alghonaim *et al.* 2025). Surface charge, chemical reactivity, and gradual, regulated, and sufficient release are characteristics that set nanomaterials apart from bulk materials (Otari *et al.* 2024). The presence of nanoparticles will increase the concentrations of related ions in the nearby soil (Chen 2018).

Joudeh and Linke (2022) attributed the action of Se in plants to the control of photosynthesis and the antioxidant defense system. The photosynthetic rate was considerably decreased by salt stress, but the foliar spray containing CuO-NPs enhanced growth and photosynthetic characteristics of maize plants (Shafiq *et al.* 2024). CuO nanoparticles can improve nutrient uptake and boost plant growth at low concentrations, but at high concentrations, they can cause phytotoxicity, which reduces growth and yield by causing oxidative stress, DNA damage, and disruption of physiological processes like photosynthesis and enzyme activity.

CuO NPs can also build up in soil and plant tissues, which could be harmful to human health because of the food chain (Ibrahim *et al.* 2022). Aside from its use in agricultural domains to treat salinity stress of maize plants, the current study sought to synthesize Se and Se/CuO NPs in doped form for the first time, utilizing extract from *Ficus religiosa* fruit. It also examined the effects of this foliar application on the plants' biochemistry, morphology, and phenolic compounds under salinity stress. Also, the phenolic profile was assessed using HPLC under different treatments.

EXPERIMENTAL

Ficus religiosa Fruit Extract

After being cleansed with distilled deionized water, the *Ficus religiosa* fruits were allowed to dry for five days at room temperature in the shade. After that, 100 mL of double-distilled deionized water and 5 g of the plant powder were heated for 20 min at 70 °C. This solution was kept at 4 °C after being further filtered *via* Whatman No. 1 filter paper.

Production of Se and CuO/Se NPs *via* Biogenesis

A total of 20 mL of the extract and 100 mL of 10 mM Na₂SeO₃ were combined to create SeNPs. The reaction took place in a darkened atmosphere with continuous magnetic stirring, and it was magnetically stirred at 1200 rpm for a full day at room temperature. Se-NPs are collected and dehydrated for a variety of studies (Soliman *et al.* 2024). To create CuO/Se NPs, 0.01 mM of copper sulfate and 1 mM of sodium selenite were utilized. The synthesis procedure was finished by heating a stock solution of both precursor salts in an equal volume of 60 mL for 10 min at 80 °C. Using a magnetic stirrer, 30 mL of plant extract was constantly stirred for 1 h at 40 °C. Black green of the bimetallic CuO/Se NP precipitates replaced the pale red color of the extract. Following a 20-min centrifugation at 10,000 rpm to collect the NPs, they were meticulously cleaned three times in double-distilled deionized water to get rid of any remaining plant organic residue. It was then dried using hot air (Alsalamah *et al.* 2023b).

Characterization of Se and Se/CuO NPs

A JEM-2100 PLUS electron microscope (JEOL, Japan) operating at 200 kV with a LaB6 source was used to analyze Se/CuO NPs utilizing transmission electron microscopy (TEM) pictures and selected area electron diffraction (SAED) patterns. The assigned active groups of the active compounds that contribute to the generation of NPs were investigated using FTIR. The crystallinity, size of the crystallites, and lattice of the generated Se and Se/CuO NPs were measured using Shimadzu equipment (Shimadzu XRD-6000 lists, SSI, Japan). Additionally, the Scherer equation, $D = 0.9 \lambda / \beta \cos \theta$, was used to determine the particle size of NPs. Then, d-spacing ($d_{hkl} = \lambda / (2 \sin \theta)$), micro-strain ($\varepsilon = \beta / 4 \tan \theta$), and dislocation density ($\delta = 1/D^2$) were calculated (Tables 1 and 2).

Effects of Spraying Se and Se/CuO NPs at Varying Concentrations on *Zea mays* L. Plants

Zea mays L. (variety Giza 321) seeds were utilized in the present investigation. A field experiment was conducted using sandy loam soil. The experiment's treatments were divided into six groups. The groups were as follows: 1: control, which was irrigated with fresh water; 2: irrigated with saline water (100 mM of NaCl); 3 and 4: spray irrigated with 100 mM of NaCl with 50 and 100 ppm of Se NPs, respectively; 5 and 6: spray irrigated with 100 mM of NaCl with 50 and 100 ppm of Se/CuO NPs. Morphology measurements included the shoot and root lengths. Chlorophyll a, b, a+b, and carotenoids of leaves, also phenol, and protein of shoots were determined, and leaves' antioxidant enzymes were all included in the biochemical studies. Additionally, phenolic compounds of the control, salinity group, and salinity group in the presence of Se NPs and Se/CuO NPs (100 ppm) were estimated by HPLC. All these analyses were conducted 90 days after sowing. Biochemical investigations and controlled growth measures were conducted to gain a

deeper understanding of the effects of these elements, which are discussed in more detail in the sections that follow.

Determination of Pigment, Protein, and Phenol Contents

One gram of fresh leaves was used to assess pigment contents (chlorophyll a, b, a+b, and carotenoids) using the method described by Lichtenthaler *et al.* (1981). To remove the water-soluble proteins, the plant shoots were dried at 60 °C until a consistent dry weight was achieved. A cone was filled with 1 g of dried shoot powder that had been finely powdered. Then, 5 mL of 2% phenol water and 10 mL of distilled water were mixed. After the reaction mixture was filtered, the filtrate was diluted with distilled water to adjust its final volume to 50 mL. Protein contents were determined in accordance with Lowry *et al.* (1951). The method outlined by Diaz and Martin (1972) was used to measure the total phenolic compounds (mg/100 g of dry weight).

Determination of Antioxidant Enzymes

Catalase

Following the manufacturer's instructions, a catalase analysis kit (Biodiagnostics Co.) was used to measure the amount of catalase present. To put it briefly, for 30 min, the cells were lysed on ice. For 3 min, cell lysates were exposed to H₂O₂. The leftover H₂O₂ was then mixed with a substrate to create N-4-antipyril-3-chloro-5-sulfonate-p-benzoquinonemonoimine, which has a maximum absorbance at 540 nm. Using a spectrophotometer (Biosystem 310 plus; Biosystems, Spain), the breakdown of H₂O₂ was measured to estimate the catalase activity. The Bradford Method was used to measure the amount of protein (Selim *et al.* 2024).

Superoxide dismutase

Measuring the SOD enzyme's suppression of nitro blue tetrazolium (NBT) photoreduction allowed for the determination of SOD activity. The reaction mixture had a final volume of 3.0 mL and contained 50 mM sodium phosphate buffer (pH7.6), 0.1 mM EDTA, 50 mM sodium carbonate, 12 mM L-methionine, 50 µM NBT, 10 µM riboflavin, and 100 µL of crude extract. Crude extract was absent from a control reaction. For 15 min at room temperature, the reaction mixture was exposed to white light to perform the SOD reaction. Following a 15-min incubation period, a spectrophotometer (Biosystem 310 plus; Biosystems, Spain), was used to measure absorbance at 560 nm. An enzyme's ability to block the photochemical degradation of NBT by 50% was referred to as one unit (U) of SOD activity (Aebi 1983).

Glutathione reductase

The sample solution was a total of 200 µL; 100 µL of 2.1 mM NADPH, 100 µL of 6 mM 5,5-dithio-bis-2-nitrobenzoic acid (DTNB), 20 µL of glutathione reductase, and 125 mM sodium phosphate (pH 7.5) and 6.3 mM EDTA were included in the 1 mL reaction mixtures. Changes in optical density were monitored at 25 °C and 412 nm.

HPLC Analysis of Phenolic Compounds

Phenolic compounds of the control, salinity group, and salinity group in the presence of Se NPs and Se/CuO NPs (100 ppm) were estimated by HPLC. An Agilent 1260 series instrument was used for HPLC analysis. Using a Zorbax Eclipse Plus C8 column

(4.6 mm x 250 mm i.d., 5 μ m), the separation was performed. At a flow rate of 0.9 mL/min, the mobile phase was made up of water (A) and 0.05% trifluoroacetic acid in acetonitrile (B). The following is the sequential linear gradient programming of the mobile phase: 82% A; 0 to 1 min; 82% A; 75% A; 75% A; 11 to 18 min; 60% A; 82% A; 18 to 22 min; 22 to 24 min; 82% A). The monitoring wavelength was 280 nm for the multi-wavelength detector. Five microliters (μ L) were the injection volume for every sample solution. At 40 $^{\circ}$ C, the column temperature was kept constant (Alsalamah *et al.* 2023a).

Analytical Statistics

Utilizing Minitab 18 (Minitab LLC, State College, PA, USA), statistical evaluation was computed. *Post hoc* analyses were conducted using Tukey's test (honest significant difference), with a significance level of $p < 0.05$ and 3 replicates ($n = 3$).

RESULTS AND DISCUSSION

Characterization of Se and Se/CuO NPs

A TEM image and SAED patterns are displayed for Se and Se/CuO NPs in Fig. 1 (a,d) and (b,e), respectively. The TEM picture of the NPs showed semispherical and aggregated spherical forms. The Se and Se/CuO NPs were found to have an average particle size of 135.17 and 75.1 nm using the ImageJ tool. Figure 1b, e also displays the SAED patterns of the Se and Se/CuO samples. The SAED patterns of this material reveal polycrystalline concentric rings with diffraction spots. A protective shell forms around the particles in the Se/CuO sample due to the high concentration of phytochemicals from plant extract, altering their size and shape (Elkady *et al.* 2025).

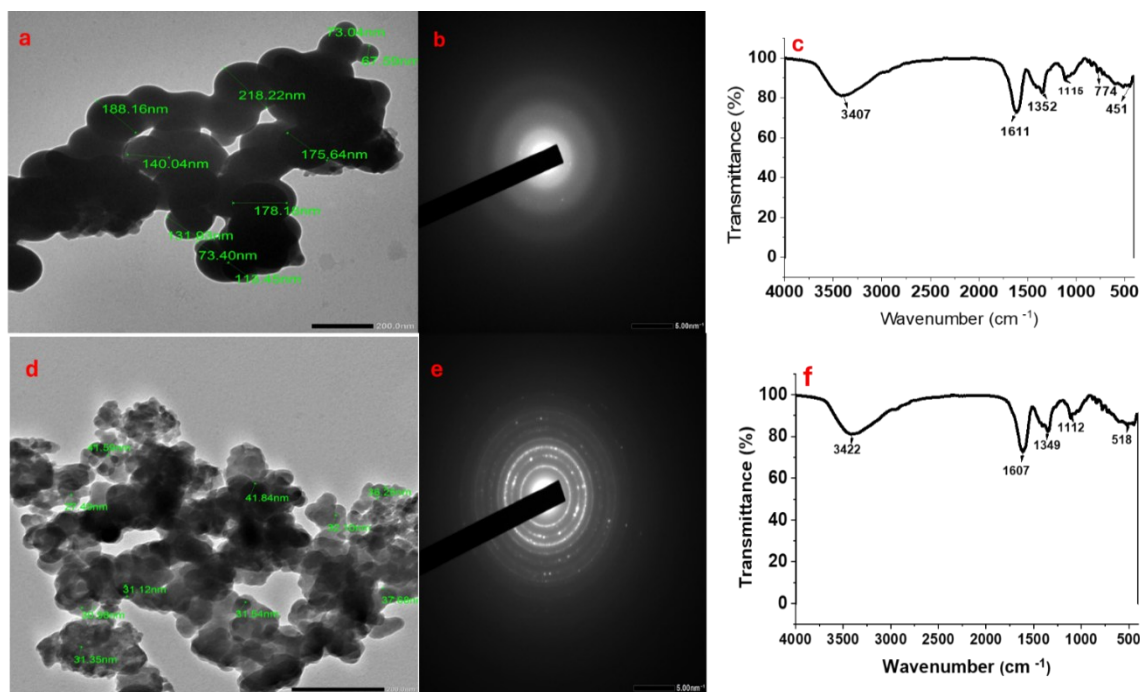


Fig. 1. TEM images (a, d), SAED patterns (b, e), and FTIR (c, f) of CuO, Se, and CuO/Se samples, respectively

The XRD patterns of the Se and Se/CuO NPs are displayed in Fig. 2a and b, respectively. The production of monoclinic CuO nanoparticles and trigonal Se nanoparticles, which matched JCPDS cards No. 80-1268 and 006-0362, respectively, was shown by the XRD pattern for Se and Se/CuO NPs (Vinu *et al.* 2021). Further supporting the Se and Se/CuO NPs' crystallinity were a few distinct peaks in the XRD pattern (Fig. 2 and Table 1). The peaks at 23.8°, 29.9°, 33.2°, 41.2°, 43.8°, 45.4°, 48.3°, 51.8°, 56.4°, and 66.1°, respectively, are identified by the hkl Miller indices as belonging to crystal planes 100, 101, 032, 110, 102, 111, 200, 414, 201, and 210 Crystalline Se (COD9008579, 9008581, and JCPDS card No. 06-362) (Safaei *et al.* 2022).

At 43.3°, 50.6°, and 74.2°, another set of 2θ peaks appeared in the XRD spectrum shown in Fig. 2; these angles correspondingly matched the Miller indices of 111, 200, and 220 Crystalline CuO (which matched JCPDS cards No. 80-1268). The phytochemicals in plant extract may be the source of additional impurity peaks. According to the Scherer equation, the average particle size of the Se and Se/CuO NPs was 16.7 and 46.2 nm, which was somewhat smaller than the TEM measurement. For smaller nanoparticles, TEM revealed a strong correlation between the size of the crystallite and the corresponding particle size (Munawar *et al.* 2024).

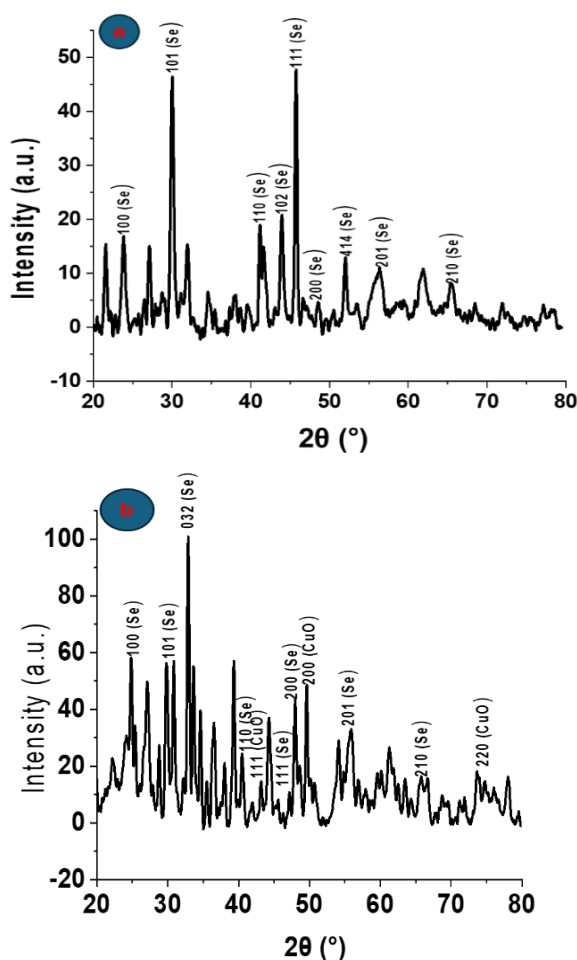


Fig. 2. XRD of Se (a) and Se/CuO NPs (b)

The fruit extract functions as a capping and stabilizing agent to produce metal NPs from the metal ions, resulting in vibrations and stretching peaks that are visible in the FTIR spectrum (Fig. 1c, f). Alcohols and phenols' O-H stretching vibrations were detected at 3407 and 3422 cm^{-1} (Demir *et al.* 2018). The C-N stretching of aromatic amine groups and the N-O symmetric stretching of nitro compounds are associated with the absorption bands at 1352 and 1349 cm^{-1} in Se NPs, and Se/CuO NPs, respectively (Rastogi and Arunachalam 2012).

Table 1. XRD Parameters for Se NPs

2 θ (°)	FWHM (β)	Size (nm)	Dislocation Density (δ)	Micro-Strain (ϵ)	d-Spacing
23.85	1.21	81.27	0.15	2.06	1.90
29.91	1.54	17.94	3.10	7.48	1.54
41.26	0.36	10.27	9.47	9.57	1.16
43.82	0.33	18.95	2.78	4.89	1.11
45.42	0.61	25.66	1.51	3.49	1.08
48.3	1.65	5.276	35.9	16.04	1.03
51.81	0.39	21.69	2.12	3.65	0.97
56.41	0.32	6.52	23.4	11.28	0.92
66.12	0.50	10.45	9.14	6.07	0.84
Average	0.76	16.749	11.94	9.97	1.17

Table 2. XRD Parameters for Se/CuO NPs

2 θ (°)	FWHM (β)	Size (nm)	Dislocation Density (δ)	Micro-strain (ϵ)	d-Spacing
24.85	1.80	4.49	49.41	35.80	1.83
29.74	1.54	5.32	35.23	25.35	1.55
40.45	0.09	93.86	0.11	1.068	1.18
43.13	0.04	185.29	0.02	0.508	1.12
47.08	0.84	10.26	9.49	8.453	1.05
48.01	0.45	19.14	2.72	4.451	1.03
49.53	0.33	25.80	1.50	3.206	1.01
55.93	1.46	6.14	26.46	12.02	0.92
65.82	0.15	59.21	0.28	1.07	0.84
73.62	0.19	52.09	0.36	1.11	0.80
Average	0.69	46.16	12.56	9.30	1.13

The spectra of nanoparticles include all of these functional groups, as shown in Fig. 1c, f. However, as mentioned, interactions between Se and CuO nanoparticles may result in pre-shift and post-shift in peaks (Thirupathi *et al.* 2024). Se and Se/CuO nanocomposites' structural and morphological properties dictate their efficacy in biological systems as mentioned in other scientific papers (Abdelhady *et al.* 2024; Rasheed *et al.* 2024). The size, shape, crystallinity, and surface chemistry of particles are meticulously designed to enhance their bioavailability, cell targeting, and therapeutic efficacy.

Shoot and Root Lengths of Maize Plants in Response to Se or Se/CuO NPs

Salinity significantly impacted several physio-biochemical characteristics, including photosynthetic activity and biomass production. Although plants responded differently to salt stress, it had detrimental cellular impacts on entire plants (Rahman *et al.* 2021). Plants attempt to increase their tolerance under stress by changing their physiological and biochemical functions. Turgor losses have frequently been seen in saline environments, which can have fatal consequences for plant growth because they create hypertonic conditions around the cell. The use of nanoparticles is essential in agriculture to prevent salt stress because it either improves antioxidant production or slows the synthesis of reactive oxygen species (ROS) (Al-Rajhi *et al.* 2022). Figure 3 shows that the salinity at 100 mM showed significant decrease in shoot and root lengths of maize plants. Se NPs at 100 ppm and Se/CuO at 50 and 100 ppm appeared to have significantly improved these morphological parameters compared to the salinity group. Depending on the plant species, age, and metal nanoparticle concentration, several studies have found that applying nanoparticles improves the quality of various crops, including citrus fruit juice (Shani *et al.* 2023), as well as cucumber, tomato, mustard, Arabidopsis, and maize crops, which helps them adapt to varying environmental conditions (Wu *et al.* 2023). Prior research demonstrated that CuO-NPs made plants more resilient to biotic and abiotic stressors (Yadav *et al.* 2023). Shoot, root, and growth were all extremely sensitive to salinity stress, which severely damaged the leaves' mesophyll area and reduced the biomass production of maize plants (Abbasi *et al.* 2015). Another study found that the application of mixed nanoparticles of Se and Zn was especially successful in bringing physiological and biochemical parameters back to levels that were close to normal in rice plants under salinity stress. These gains included increases of 46.3% in plant height, and 70.5% in root length (Mishra *et al.* 2025).

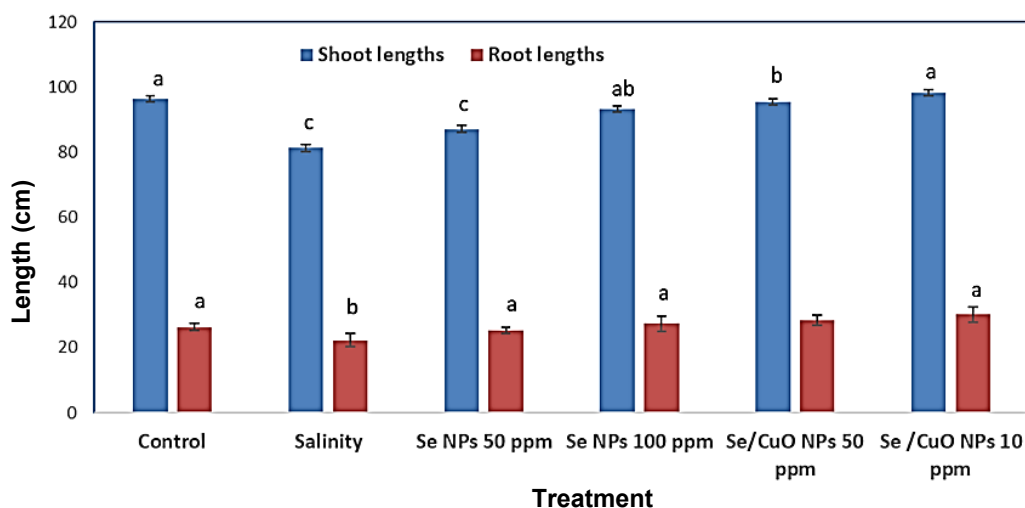


Fig. 3. Shoot and root lengths of maize plants under salinity stress in response to Se and Se/CuO NPs

Pigments, Protein, and Phenol Contents

Data in Table 3 indicate that salinity caused a significant decrease in pigment and protein content, but there appeared to be an increase in phenol contents. Through increasing the rate of photosynthesis and subsequently lengthening the shoots and roots, foliar application of Se and Se/CuO NPs is essential for improving vegetative growth. Salinity-induced oxidative stress has been linked to adverse effects on plant growth, physio-biochemical processes, anatomical features, metabolic processes, and enzyme activity. The thickness of cell walls of resistant types is greatly increased by high salt concentrations, which also increases chloroplasts and intercellular gaps (Arif *et al.* 2020).

Table 3. Effect of Se and Se/CuO on Pigments, Protein, and Phenols Contents of Maize Plants Under Salinity Stress

Treatments	Chlorophyll a (mg/g Fresh Weight)	Chlorophyll b (mg/g Fresh Weight)	Chlorophyll a+b (mg/g Fresh Weight)	Carotenoids (mg/g Fresh Weight)	Protein (mg/g Dry Weight)	Phenols (mg/g Dry Weight)
Control	1.78 ± 0.05c	3.48 ± 0.21c	5.27 ± 0.32c	0.68 ± 0.01c	9.62 ± 0.36d	0.101 ± 0.003c
Salinity	0.92 ± 0.03d	2.54 ± 0.11d	3.46 ± 0.19d	0.47 ± 0.04d	8.94 ± 0.16e	0.197 ± 0.011a
Se NPs 50 ppm	1.94 ± 0.01b	3.91 ± 0.13b	5.86 ± 0.12b	0.59 ± 0.03b	9.67 ± 0.25d	0.155 ± 0.008b
Se NPs 100 ppm	2.05 ± 0.05b	3.98 ± 0.06b	6.05 ± 0.26b	0.63 ± 0.05b	11.36 ± 0.65b	0.148 ± 0.013b
Se/CuO NPs 50 ppm	2.40 ± 0.04a	4.50 ± 0.24a	6.91 ± 0.39a	0.71 ± 0.07a	10.34 ± 0.46c	0.115 ± 0.009c
Se /CuO NPs 10 ppm	2.65 ± 0.08a	4.65 ± 0.16a	7.32 ± 0.14a	0.80 ± 0.09a	13.18 ± 0.21a	0.108 ± 0.008c
LSD	0.316	0.262	0.531	0.122	0.321	0.019

Se/CuO NPs at 100 ppm caused an increased value of chlorophyll a, b, a+b, carotenoids and protein contents by 188.0, 83.1, 111.6, 70.2, and 47.4% compared to the salinity group. Another study by Vishwakarma *et al.* (2018) demonstrated that the total protein content distributed in tomato shoots and roots was considerably enhanced by foliar treatment with CuO-NPs.

Antioxidant Enzymes

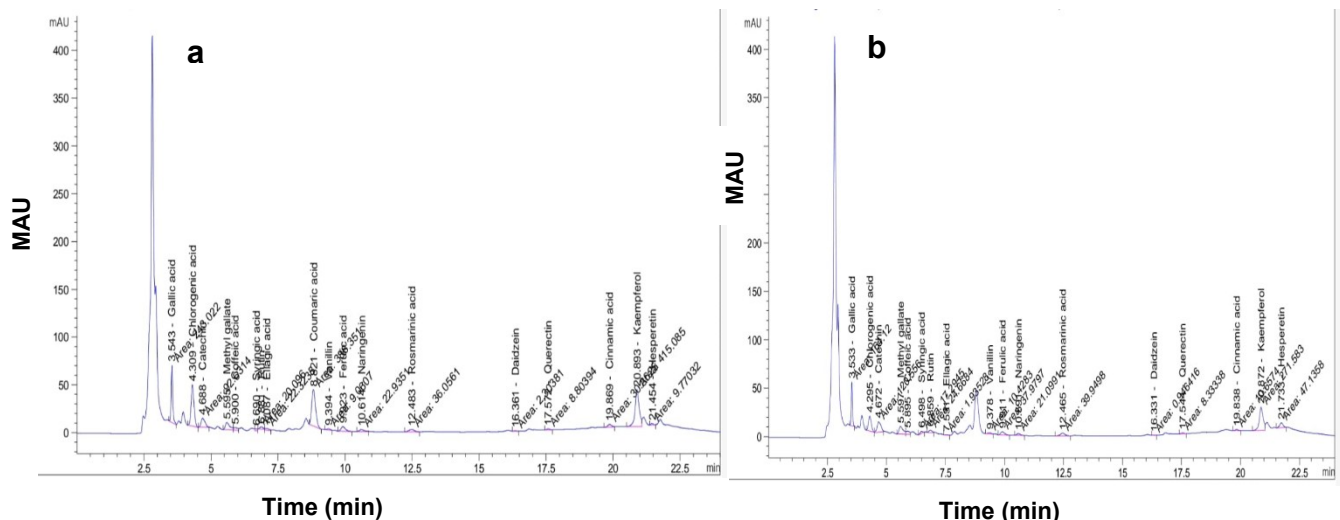
Data in Table 4 indicate that both salinity and Se or Se/CuO nanoparticles caused a significant increase in antioxidant enzymes (catalase, superoxide dismutase, and glutathione reductase). Se/CuO NPs at 100 ppm showed the higher activity of catalase, superoxide dismutase, and glutathione reductase enzymes of 68.1, 66.3, and 245.2 mmol/g, respectively, compared to the control group. Through the enhancement of their antioxidant system, nanoparticles help plants become more resilient to oxidative stress. Antioxidative defense systems, such as SOD, and catalase, are activated in response to oxidative stress caused by nanoparticles. Low oxidative stress caused by low nanoparticle concentrations in plants results in a reduction in antioxidant activity (Sharma *et al.* 2019).

Table 4. Effect of Se and Se/CuO on Antioxidant Enzymes (Catalase, Superoxide Dismutase, and Glutathione Reductase) of Maize Plants Under Salinity Stress

Treatments	Catalase (U/g)	SOD (U/g)	Glutathione Reduced (mmol/g)
Control	44.23 ± 0.68f	79.47 ± 0.33e	12.53 ± 0.21d
Salinity	49.67 ± 0.44e	89.60 ± 0.25d	22.55 ± 0.35c
Se NPs 50 ppm	56.87 ± 0.18d	115.30 ± 0.54c	36.10 ± 0.15b
Se NPs 100 ppm	61.23 ± 0.19c	122.12 ± 1.02b	39.02 ± 0.41a
Se/CuO NPs 50 ppm	69.30 ± 0.42b	126.97 ± 1.03a	40.80 ± 0.56a
Se /CuO NPs 100 ppm	74.36 ± 0.74a	132.14 ± 2.12a	43.25 ± 0.45a
LSD	3.15	6.45	4.36

Phenolic and Flavonoid Compounds of Maize Plants Under Different Treatments

Table 5 and Fig. 4 indicate the impact of salinity in the presence or absence of Se or Se/CuO NPs at 100 ppm on phenolic compounds of maize plants using HPLC. Salinity at 100 mM caused a decrease in gallic acid, chlorogenic acid, ellagic acid, ferulic acid, naringenin, daidzein, Quercetin, cinnamic acid, and kaempferol contents. In the same trend, under salinity, phenolic compounds concentration, such as gallic, syringic, and p-coumaric acids, vanillic, and caffeic acids, increased in grape plants, which have played a significant part in salinity tolerance (Mohammadkhani 2018). The highest total phenolic compound content in *Glaux maritima* was found to be between 50 and 100 mM of salinity stress (Pungin *et al.* 2023).



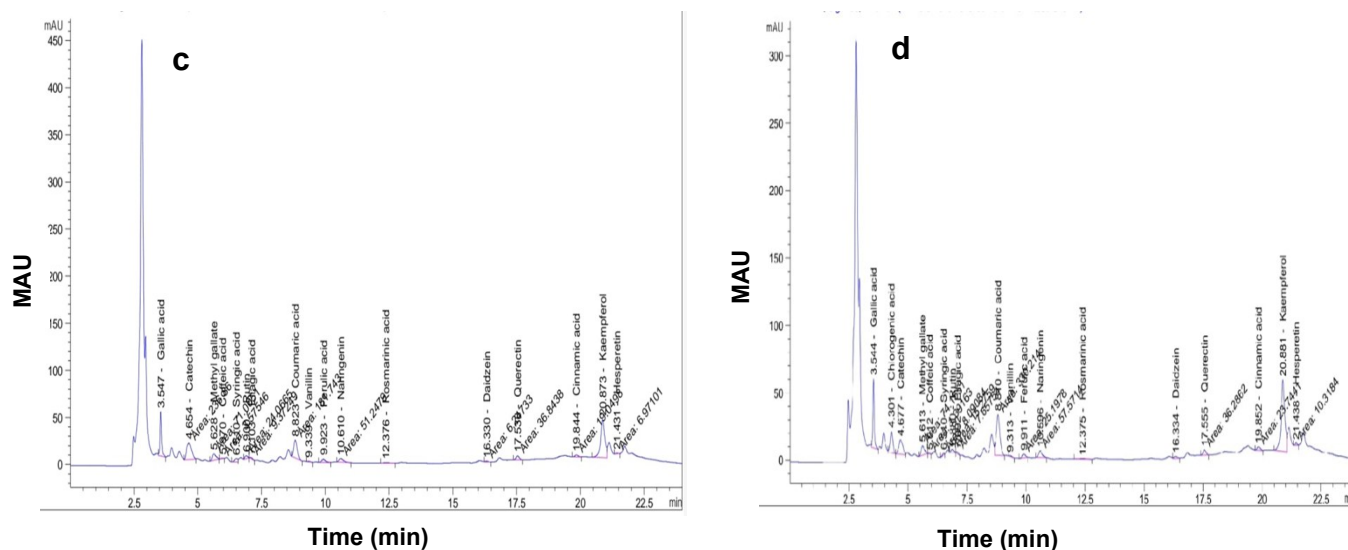


Fig. 4. HPLC for the detection of phenolic compounds in *Zea mays* leaves at different treatments: (a) Control, (b) Salinity group, (c, d) Salinity group treated with 100 ppm of Se and Se/CuO NPs

Table 5. Effects of Se and Se/CuO NPs on Phenolic Compounds on *Zea mays* L. Based on HPLC Analysis

Phenolics	Control	Salinity (100 mM)	Se (100 ppm)	Se/CuO (ppm)
	Conc. (µg/g)			
Gallic acid	880.48	594.61	736.57	796.45
Chlorogenic acid	2272.83	888.28	0.00	901.84
Catechin	1075.33	1399.50	2749.64	1560.78
Methyl gallate	255.77	273.23	201.27	210.19
Caffeic acid	87.84	103.16	10.70	4.17
Syringic acid	42.45	55.36	97.61	25.90
Rutin	146.09	179.32	174.95	111.05
Ellagic acid	126.91	11.00	53.28	43.53
Coumaric acid	705.43	0.00	331.84	602.93
Vanillin	15.33	17.65	6.10	8.11
Ferulic acid	152.07	109.83	114.81	72.87
Naringenin	110.87	101.99	247.73	278.30
Rosmarinic acid	160.18	177.48	31.86	47.33
Daidzein	6.59	2.71	17.86	34.80
Quercetin	57.01	53.96	238.57	234.96
Cinnamic acid	32.86	11.53	20.62	25.70
Kaempferol	2127.93	1392.27	2369.70	2992.54
Hesperetin	23.10	111.45	16.48	24.40

In the present study, Se NPs at 100 ppm had the highest values of catechin, syringic acid, and quercetin. Se/CuO NPs at 100 ppm appeared to have the highest values of

naringenin, daidzein, kaempferol, and hesperetin. To fend off ROS, plants employ their receptors to identify danger and trigger a natural defense mechanism. One of these defense mechanisms is the buildup of specific protective secondary metabolites, including terpenes, alkaloids, and phenolic compounds (PCs). Particularly, PCs function as strong antioxidants and are necessary for the plant to survive salt stress. The plants' endurance, competitiveness, tenacity, and survival against salinity stress are ensured by enhanced PC synthesis (Reetu *et al.* 2023). Because phenolic compounds have potent antioxidant activity, their accumulation is one of the nonenzymatic antioxidant defense mechanisms that shield plants from harm caused by ROS. In another study on garlic bulbs, it was found that the nanoparticles effectively increased the levels of flavonoids and total phenolic compounds, while Se NPs promoted these substances, particularly at 0.5 ppm (El-Saber 2021).

CONCLUSIONS

1. Plant metabolism and resistance to salinity stress were improved by Se and Se/CuO nanoparticle (NPs).
2. Significantly, the morphological characteristics (shoot and root lengths) and biochemical traits (pigment contents, protein, and carbohydrates) of the 100 ppm of Se/Cu group under salinity stress were similar to those of the control group.
3. According to the current analysis, plant development was negatively impacted by salinity. In fact, the results demonstrated that salinity had a detrimental effect on a variety of physiological markers, such as total protein, total phenolic content, carotenoid content, and total chlorophyll content.
4. The foliar spray of selenium and selenium/copper oxide nanoparticles helped maize plants overcome the salt stress by enhancing the previously mentioned metrics. Therefore, the negative effects of salt may be lessened by applying Se and Se/CuO-NPs topically at particular crop phenological stages. To ascertain the concentration of nanoparticle suspensions and their use at various phenological stages, further investigation is necessary.

Abbreviations List

Phenolic compounds (PCs), Superoxide dismutase (SOD), Nanoparticles (NPs), Salinity stress (SS), Reactive oxygen species (ROS)

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