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# Potassium Silicate Decreases Nickel Induced Oxidative Stress by Improving Nutrients Uptake and Antioxidant Defense System in Sunflower

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## ABSTRACT

Nickel stress reduces nutrient uptake, leading to oxidative stress and inhibiting plant growth and development. There is insufficient research on the detrimental effects of nickel stress on sunflowers. This study aimed to evaluate the effects of potassium silicate at concentrations of 50 and 100 mg L-1 on the morphophysiological, nutritional, and biochemical characteristics of sunflower plants subjected to nickel stress at levels of 100 and 200 mg L-1. This experiment utilized a completely randomized design with a factorial approach, incorporating three biological replications. The nickel stress negatively impacted plant growth parameters, with the most significant effects observed at high nickel concentrations (200 mg L-1). Under significant nickel stress, potassium silicate (K2SiO3) at a concentration of 100 mg L-1 enhanced plant morphophysiological characteristics, while 50 mg L-1 exhibited non-significant variation. The primary reason may be attributed to enhanced total chlorophyll, nitrogen, phosphorus, and potassium uptake from the soil, with increases of 13.33%, 24.04%, 7.40%, and 40.96%, respectively, following the application of K2SiO3 at a concentration of 50 mg L-1 under nickel stress at 200 mg L-1. This may be linked to a reduction in oxidative stress, encompassing electrolyte leakage, malondialdehyde, and hydrogen peroxide by 23.06%, 10%, and 25.38% respectively under nickel stress. The reduction in oxidative stress may result from enhanced antioxidant activities, specifically superoxide dismutase and catalase, which increased by 31.67% and 7.90%, respectively, under nickel stress. The potassium silicate at 50 mg L-1 was considered as best dose in improving plant growth, nutritional, physiological and biochemical aspects of sunflower under severe nickel stress.

## INTRODUCTION

Soil contamination by industrial waste represents a critical environmental concern, given that soil microbial activity and diversity exhibit high sensitivity to both organic and inorganic pollutants, including heavy metals (Ishaq et al., 2024). Heavy metals are classified as contaminants because of their toxicity, bioaccumulation, persistence, and resistance to degradation (Siddique et al., 2018). Heavy metals emitted by industries are classified as environmental pollutants due to their toxic effects on flora, fauna, and humans. They disrupt physiological processes in plants, including photosynthesis, gas exchange, and nutrient uptake, leading to decreased growth, reduced dry matter accumulation, and lower yields (Siyar et al., 2020). Soil contamination by heavy metals represents a global issue, leading to decreased agricultural productivity and significant health risks upon entry into the food chain (Aioub et al., 2019). Nickel, identified in 1975 as a component of the urease enzyme, is an essential micronutrient for plant growth and plays a critical role in nitrogen metabolism in higher plants (Naveed et al., 2020). The environmental concentration of nickel is rising as a result of its application in commercial and industrial sectors. The use of industrial waste in agricultural regions results in the introduction of trace elements into the environment, which subsequently elevates nickel concentrations in the soil (Syed et al., 2023). The anthropogenic release contributes to an increased concentration of nickel in plants and the food chain (Ishaq et al., 2024). Nickel, a crucial element in seed and vegetative tissues, poses a significant health risk due to its impact on various physiological and biochemical processes (Naveed et al., 2020). Common symptoms include respiration issues, chlorosis, and inhibited photosynthesis. Nichol can exist in soil in multiple forms, including precipitates, complexes absorbed on cation exchange sites, organic cation surfaces, free ions, or as complex chelated metals in soil solutions (Hafeez et al., 2024; Saif and Khan, 2018).

Plants require a range of major and trace nutrients for healthy growth, with the ultimate source being the geological mineral assemblage within a soil (Mona et al., 2021; Sarker et al., 2018). Soil minerals play a crucial role in controlling K and N (as ammonium) within the soil solution. Potassium containing fertilizers are the dominant source of potassium for plant growth, but it is considered inert on the timescale of crop production due to its slow dissolution rate and low K yields (Ozlu et al., 2019). Other widely occurring potassium silicate minerals include micas, which have been known to act as sources of K through cation exchange and weathering reactions (Nazim et al., 2024). Globally, inputs of potassium to soils from all sources are much less than the amount removed through the harvesting of crops. Nutrient balance studies show that if this deficit was an accurate representation of the need for potassium to support crop growth, then the requirement for supplementation with potassium would be far greater than the need for P or N, which more often are in balance (Nelkner et al., 2019; Rawat et al., 2022). Farmers must pay high associated transport and distribution costs, as well as the high soluble nature of K from sources like KCl, which can be rapidly lost through leaching (Rajadurai et al., 2022). To supply the K required for crop growth, it is important to consider novel inputs in addition to conventional sources, given constraints on price and availability. Climate changes, such as altered precipitation and temperature systems, have negatively affected crop quantity and yields.

Silicon is helpful to plants by reducing the absorption of high levels of nutrients, preventing tissue damage, and playing a role in ethylene inhibition (Nisar et al., 2022). It also helps crops maintain freshness longer with better appearance. According to Rizwan et al., (2019), the grain yield of rice was improved with the application of Si fertilizer. Potassium is essential for basic physiological functions of plants, such as protein synthesis, sugars and starch formation, and cell division, while silicon stimulates plant growth and enhances productivity across crops (Ahmed et al., 2024; Massoud et al., 2024). Understanding the protective role of potassium silicate against environmental stress remains limited. This will be crucial in formulating effective ways to improve sunflower crop resilience and productivity in challenging conditions. Therefore, we hypothesized that soil application of potassium silicate could improve nutrients uptake and improve sunflower growth under nickel contaminated soil. The main objective of this study was (i) to access the different doses of nickel on sunflower growth aspects (ii) to access the potential of potassium silicate to decrease oxidative stress by boosting antioxidant enzymes and improve sunflower growth.

## MATERIAL AND METHODS

The experiment took place in the University of Agriculture Peshawar, during the 2022-2023 academic session. The experiment was conducted to facilitate a comparative analysis of sunflower varieties under nickel stress conditions. Sunflower seeds were sourced from the oil seeds section at the Ayub Agriculture Research Institute (AARI) in Faisalabad. No permission was required for the allocation of seeds from the institute. The seeds underwent surface sterilization using a 0.1% sodium hypochlorite solution, followed by three rinses with double distilled water. Earthen pots measuring 22 cm by 20 cm were filled with 5 kg of clayey, loamy soil, and a hole of equal size was marked at the bottom of each pot. Each pot contained ten seeds, which were then covered with a thin layer of sand. Nutrients were applied at the time of sowing, with initial levels of N: 19 mg, P: 19 mg, and K: 12 mg. Nickel concentrations of 100 and 200 mg $L^{-1}$ were prepared and applied to sunflower roots after one week of germination. Potassium silicate solutions at concentrations of 50 and 100 mg $L^{-1}$ were prepared and applied in soil after three days of stress imposition. Foliar application of potassium silicate was administered to the leaves of all pots for both varieties. The experiment utilized a completely randomized design (CRD) featuring a factorial arrangement with three biological replications. Factor one is cobalt stress, while factor two is potassium silicate. All agronomic practices were conducted during the experiment.

### Morphological Aspects

In this study, a 10-day stress application, followed by irrigation to loosen soil and uproot seedlings to prevent root damage. Morphological attributes like root and shoot length, fresh and dry weight were assessed. Uprooted seedlings were washed with tap water, dried in the shade, sun dried for three days, and oven-dried at 65°C until a constant weight was achieved. The effective dose was determined and used for physiological and biochemical analysis. The results were used to determine the optimal treatment for sunflower seedlings.

### Plant N, P and K Uptake from Soil

The plant N uptake of shoot and root was estimated by Kjeldahl method after oven drying at 105 °C (Chapman and Pratt, 1962). The spectrophotometer method was used to analyzed plant P uptake of shoot and root (Chapman and Pratt, 1962). The flame photometer method was used to analyzed plant K uptake of shoot and root (Chapman and Pratt, 1962).

### Total Chlorophyll

Fresh leaves were crushed in 80% acetone, which included purification and centrifugation at 13,000 rpm for 20 minutes at 4°C (Arnon, 1949). The remainder of the mixture was then separated, and the absorbance of the sample was measured using a UV-visible spectrometer at wavelengths of 665, 649, and 470 nm.

### Biochemical Analysis

Electrolyte leakage (EL) was assessed by washing leaves with distilled water, followed by drying and placing them in test tubes. Following 180 minutes of incubation at 25°C, the mixture underwent autoclaving for 15 minutes (Bajji et al., 2002). The hydrogen peroxide content was calculated from maize leaves by extracting and homogenizing fresh leaves with TCA, centrifuging, and measuring the absorption using a spectrophotometer at a wavelength of 390 nm (Nakano and Asada, 1981). The Davey et al. (2005) method was used to determine malondialdehyde levels. Fresh plant leaves and triturate were mixed with trichloroacetic acid, centrifuged, and incubated for 60 minutes. The samples were then cooled, centrifuged, and analyzed using a spectrophotometer at 532 and 600 nm wavelengths. Fresh leaves were ground for antioxidant enzyme extraction, homogenized, centrifuged, and supernatant determined for enzymatic antioxidants. Superoxide dismutase activity was assayed using method (Foster and Edwards, 1980). The catalase activity was measured in fresh leaves using a modified method by (Aebi, 1984), which involved adding enzyme solution, phosphate buffer, and heating, and checking absorbance.

### Statistical Analysis

The collected data was rearranged using MS excel and then analyzed using analysis of variance (ANOVA). The mean values was analyzed using tukey HSD.test (Statistics, 2013). The data was visualized by using R programming software.

## RESULTS

### Growth and Yield aspects

Nickel (Ni) stress significantly decreased the growth and yield aspects, including shoot length (SL), root length (RL), shoot fresh weight (SFW), shoot dry weight (SDW), root fresh weight (RFW), and root dry weight (RDW) of sunflower ($p < 0.05$). Among Ni treatments, the most pronounced decrease was noticed in Ni at 200 mg $L^{-1}$, followed by 100 mg $L^{-1}$, as shown in the table. It was noticed that Ni at 200 mg $L^{-1}$ significantly decreased SL by 46.68%, RL by 31.43%, SFW by 26.23%, SDW by 27.14%, RFW by 39.04%, and RDW by 28.54% as compared to control as shown in Table 1. The application of potassium silicate ($K_2SiO_3$) at 100 ml $L^{-1}$ gave the best response in improving growth and yield aspects of sunflower; however, it showed non-significant variation with 50 ml $L^{-1}$ under nickel stress ($p > 0.05$). Under Ni200, the $K_2SiO_3$ at 50 ml $L^{-1}$ significantly improved SL, RL, SFW, SDW, RFW, and RDW by 28.74%, 18.80%, 14.53%, 10.26%, 51.98%, and 5.75% as compared to non-$K_2SiO_3$ nickel stress.

**Table 1**

*The growth and yield aspects of sunflower*

| Ni | $K_2SiO_3$ | SL | RL | SFW | SDW | RFW | RDW |
|---|---|---|---|---|---|---|---|
| Ni0 | $K_2SiO_3$0 | 37.83±1.47b | 6.261.470.04ab | 9.890.040.08ab | 6.150.080.04ab | 5.670.040.13c | 4.220.130.21a |
| Ni1 | $K_2SiO_3$0 | 26.39±1.02c | 5.141.020.1c | 8.410.10.14c | 4.980.140.1d | 5.290.10.1c | 3.170.10.04cd |
| Ni2 | $K_2SiO_3$0 | 20.17±0.78d | 4.290.780.21d | 7.290.210.18d | 4.480.180.1e | 3.460.10.21d | 3.010.210.06d |
| Ni0 | $K_2SiO_3$1 | 42.69±0.85a | 6.530.850.07a | 10.280.070.09a | 6.330.090.06a | 6.740.060.07a | 3.550.070.02b |
| Ni1 | $K_2SiO_3$1 | 35.15±1.37b | 5.931.370.11b | 9.490.110.16b | 5.750.160.11c | 6.110.110.12b | 3.380.120.04bc |
| Ni2 | $K_2SiO_3$1 | 25.97±1.01c | 5.11.010.1c | 8.350.10.14c | 4.940.140.1d | 5.250.10.1c | 3.190.10.09cd |
| Ni0 | $K_2SiO_3$2 | 43.28±0.87a | 6.580.870.07a | 10.350.070.09a | 6.380.090.06a | 6.780.060.07a | 3.570.070.02b |
| Ni1 | $K_2SiO_3$2 | 37.18±1.45b | 6.11.450.12b | 9.710.120.16b | 5.910.160.11bc | 6.290.110.12b | 3.430.120.03bc |
| Ni2 | $K_2SiO_3$2 | 27.48±1.07c | 5.241.070.1c | 8.550.10.14c | 5.090.140.1d | 5.40.10.11c | 3.150.110.04cd |

Data is average of three means ± standard error. Common lettering showed non-significant difference among means.

### Total Chlorophyll

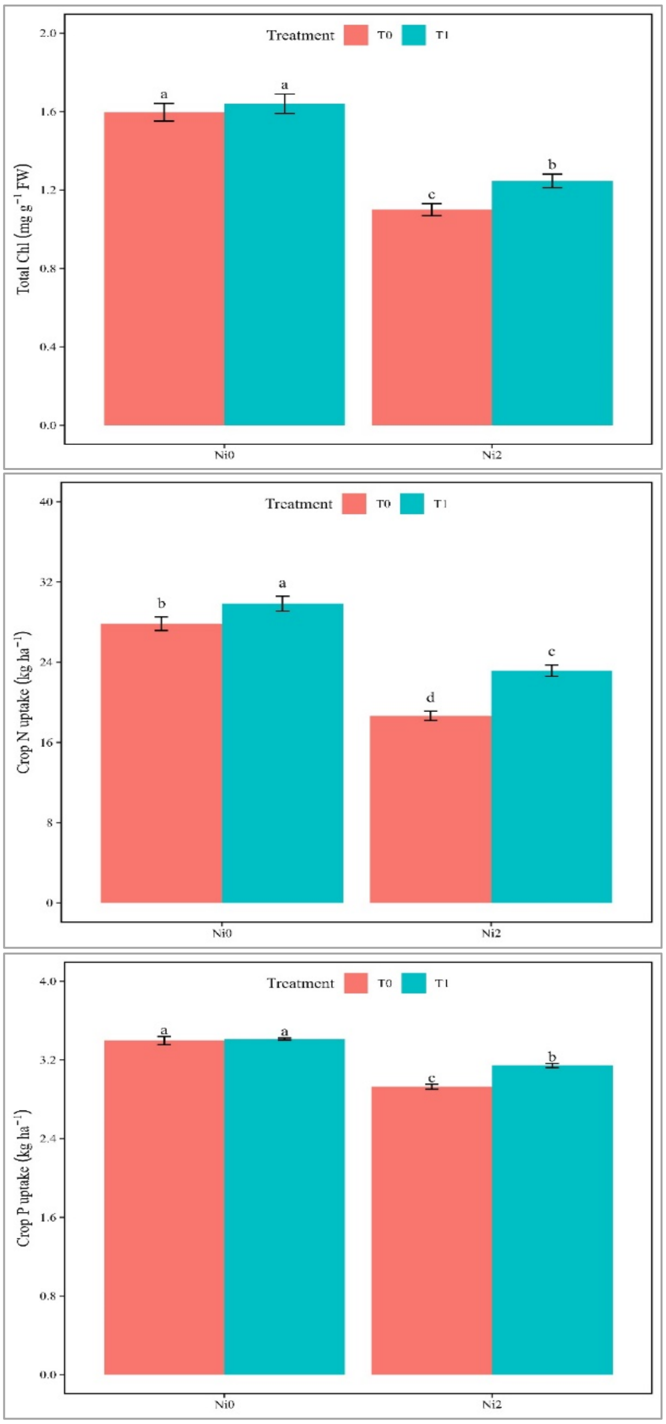
The total chlorophyll (chl) was significantly affected under severe nickel (Ni) stress ($p < 0.05$). It was noticed that the total chl was significantly decreased by 31.11% under Ni at 200 mg $L^{-1}$ as compared to control as shown in Fig. 1. The application of potassium silicate ($K_2SiO_3$) gave the best response in improving biosynthesis of chl of sunflower under nickel stress ($p > 0.05$). The $K_2SiO_3$ at 50 ml $L^{-1}$ significantly improved the biosynthesis of chl by 13.13% under Ni stress at 200 mg $L^{-1}$ over plants grown under untreated Ni stress conditions.

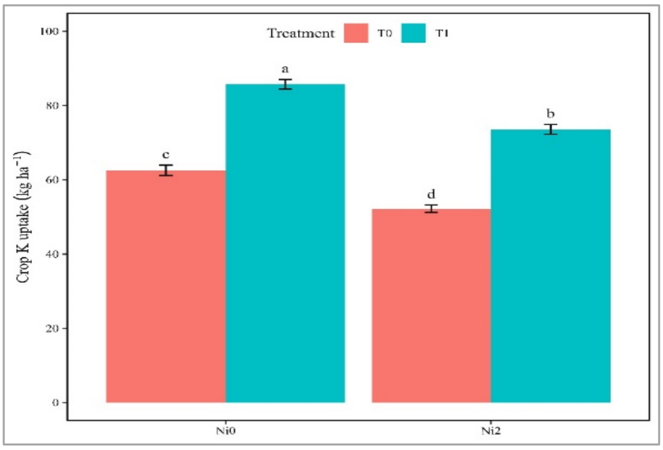
### Plant N, P, and K uptake from Soil

The plant nitrogen (N), phosphorus (P), and potassium (K) uptake from soil was significantly affected under severe nickel (Ni) stress ($p < 0.05$). It was noticed that the N, P, and K uptake was significantly decreased by 32.93%, 13.84%, and 16.60% under Ni at 200 mg $L^{-1}$ as compared to control as shown in Fig. 1. The application of potassium silicate ($K_2SiO_3$) gave the best response in improving uptake of nutrients of sunflower ($p > 0.05$). The $K_2SiO_3$ at 50 ml $L^{-1}$ significantly improved the uptake of these nutrients (N, P, and K) by 24.04%, 7.40%, and 40.96% under Ni stress at 200 mg $L^{-1}$ over plants grown under untreated Ni stress conditions.

**Figure1**

*Effect of potassium silicate ($K_2SiO_3$) on total chlorophyll, crop nitrogen (N), phosphorus (P), and potassium (K) of sunflower under nickel stress (Ni). Data is average of three means ± standard error. Common lettering showed non-significant difference among means.*



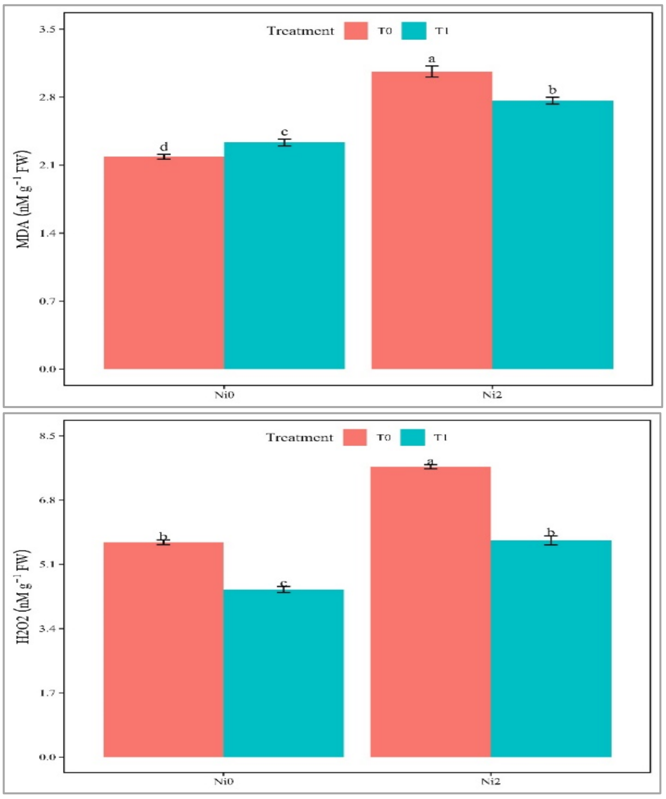



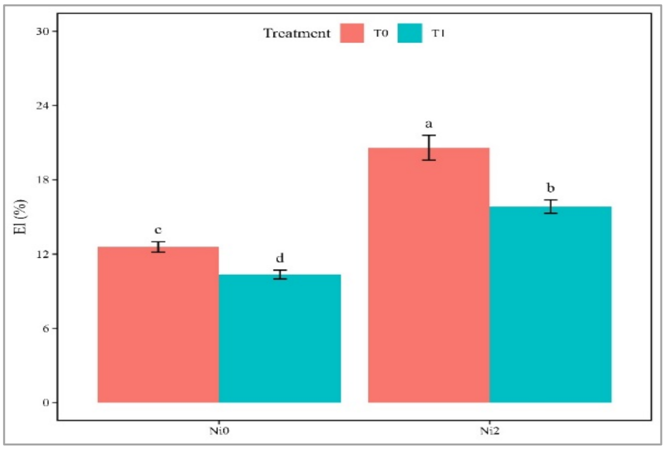

### Oxidative Stress Markers

The nickel (Ni) stress significantly increased electrolyte leakage (EL), malonaldeahyde (MDA), and hydrogen peroxide ($H_2O_2$) of sunflower ($p < 0.05$). The Ni at 200 mg $L^{-1}$ significantly increased EL by 63.69%, MDA by 40.09%, and H2O2 by 35.19% as compared to control as shown in Fig. 2. The application of potassium silicate ($K_2SiO_3$) gave the best response in decreasing these oxidative stress markers of sunflower ($p > 0.05$). The K2SiO3 at 50 ml $L^{-1}$ significantly decreased EL by 23.06%, MDA by 9.79%, and $H_2O_2$ by 25.38% as compared to non-$K_2SiO_3$ Ni stress conditions.

**Figure 2**

*Effect of potassium silicate ($K_2SiO_3$) on malonaldeahyde (MDA), hydrogen peroxide ($H_2O_2$), and electrolyte leakage (EL) of sunflower under nickel stress (Ni). Data is average of three means ± standard error. Common lettering showed non-significant difference among means.*



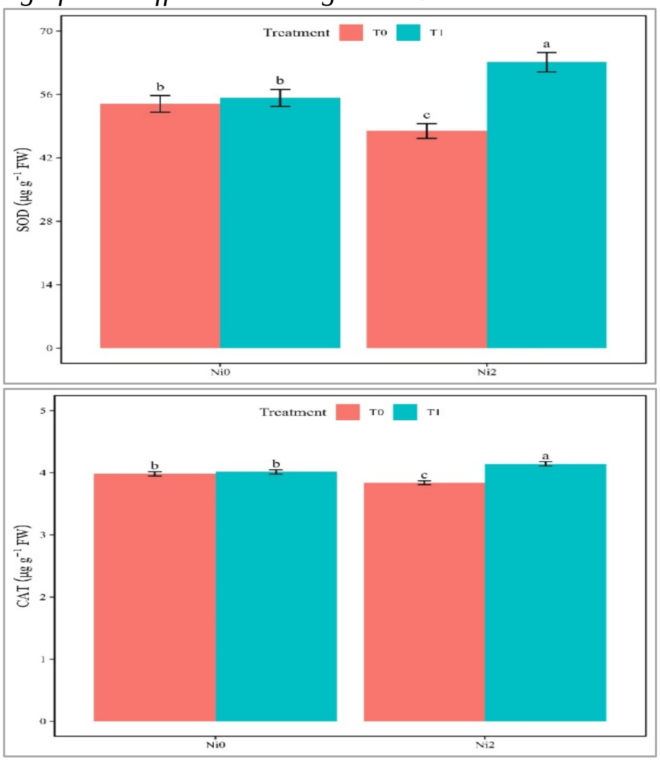



## Antioxidant enzymes

The antioxidant enzymes including superoxide dismutase (SOD) and catalase (CAT) were significantly affected under severe nickel (Ni) stress ($p < 0.05$). It was noticed that the SOD and CAT were significantly decreased by 11.08% and 3.60% under Ni at 200 mg $L^{-1}$ as compared to control as shown in Fig. 3. The application of potassium silicate ($K_2SiO_3$) gave the best response in improving activities of these antioxidant enzymes of sunflower under nickel stress ($p > 0.05$). The $K_2SiO_3$ at 50 ml $L^{-1}$ significantly improved the activities of SOD by 31.67% and CAT by 8.90% under Ni stress at 200 mg $L^{-1}$ over plants grown under untreated Ni stress conditions.

**Figure 3**

*Effect of potassium silicate ($K_2SiO_3$) on superoxide dismutase (SOD), and catalase (CAT) of sunflower under nickel stress (Ni). Data is average of three means ± standard error. Common lettering showed non-significant difference among means.*

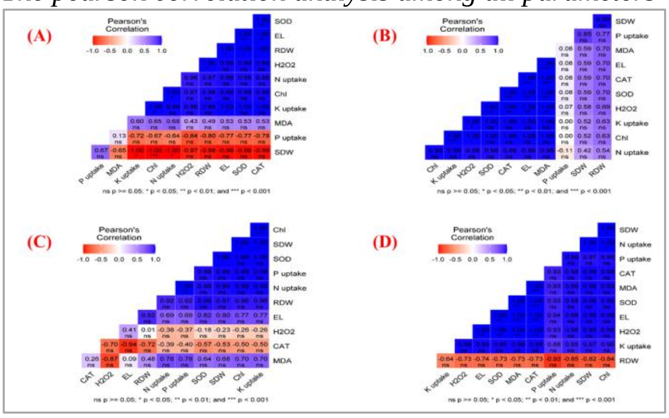



## Correlation Analysis

The Pearson correlation analysis can examine the interactions between physiological and biochemical traits of sunflowers under different treatments, control (A), potassium silicate (B), nickel stress (C), and potassium silicate under nickel stress (D) as shown in Fig. 4. Under control conditions (A), positive correlations were seen between P uptake and K uptake ($r = 0.67$, $p < 0.05$) and between P uptake and RDW ($r = 0.96$, $p < 0.001$). However, K uptake was strongly negatively correlated with malondialdehyde (MDA) ($r = -1.00$, $p < 0.001$) and with chlorophyll content (Chl) ($r = -0.77$, $p < 0.01$). In the potassium silicate treatment (B), P uptake was positively correlated with RDW ($r = 0.99$, $p < 0.001$). However, negative correlations between K uptake and hydrogen peroxide (H2O2) appeared to show that it has the potential to reduce oxidative damage ($r = -0.72$, $p < 0.05$). The correlation between P uptake and RDW became weakened under nickel stress (C), while K uptake continued to appear negatively correlated with H2O2 ($r = -0.70$, $p < 0.05$). Furthermore, the negative correlations were found between K uptake and other enzyme activities such as superoxide dismutase (SOD) ($r = -0.50$, ns) and catalase (CAT) ($r = -0.53$, ns). In nickel stress conditions, when potassium silicate was used (D), the positive correlation between P uptake and RDW enhanced ($r = 0.99$, $p < 0.001$); that means potassium silicate played its protective role against nickel-induced stress conditions. K uptake maintained its strong negative correlation with H2O2 ($r = -0.73$, $p < 0.05$) and CAT ($r = -0.73$, $p < 0.05$), suggesting its role in the regulation of oxidative stress pathways.

**Figure 4**

*The pearson correlation analysis among all parameters*




## DISCUSSION

The soil application is a technique that promotes growth and reduces the adverse impacts of heavy metal stress in agriculture (Zhu et al., 2022). Potassium silicate ($K_2SiO_3$) serves as a biosimulant for plants, providing potassium and soluble silicon, essential for improving crop production (Nazim et al., 2024). Silicon enhances plant resilience to metal stress by decreasing sodium ion absorption and facilitating potassium ion uptake in the leaves (Rachappanavar et al., 2024). According to Ali et al. (2023), this leads to improved root architecture, enhanced plant growth, vertical leaf orientation, optimized photosynthetic efficiency, and superior water regulation. Potassium is essential for plant growth, yield enhancement, and quality improvement, and it also mitigates the adverse effects of nickel stress in plants (Nazim et al., 2024; Rawat et al., 2022). There we hypothesized that potassium silicate could mitigate the negative effects of nickel toxicity in sunflower.

This research examined the impact of $K_2SiO_3$ on nutrients uptake of sunflower seedlings. The $K_2SiO_3$ function as an adsorbent, mitigating $NH_3$ volatilization during composting, enhancing substrate properties, increasing nutrient content, and improving the growth aspects of sunflower under nickel stress (da Silva and de Mello Prado, 2023; Nazim et al., 2024; Zhang et al., 2022). The introduction of a $K_2SiO_3$ resulted in decreased root dry weight and reduced height of sunflower seedlings, which can be associated with elevated levels of total nitrogen and total potassium relative to commercial substrate. Nitrogen availability is directly associated with plant growth, and soil nitrogen levels typically fluctuate, increasing and subsequently decreasing with rising pH levels (Mansour et al., 2021). The presence of phosphorus in the substrate significantly influences chlorophyll levels in sunflower seedlings, as increased phosphorus availability enhances plant uptake, facilitating crop development (Sarker et al., 2018). The heavy metal content of the substrate influences chlorophyll levels in plants, but no significant differences were observed in aboveground nickel content across the treatments. Hafeez et al. (2024) found that enrichment coefficients for Ni were greater in the belowground components compared to the aboveground components of the plant. The transport of heavy metals in the substrate exhibited a positive correlation with substrate pH, which increased following the addition of potassium silicate (Bai et al., 2020; Rachappanavar et al., 2024).

Our findings indicated that Ni concentrations were greater in the belowground sections compared to the aboveground sections, attributable to the propensity for heavy metals to accumulate predominantly in the plant's lower regions (Gonzalez-Sanchez et al., 2019). An increase in potassium silicate concentration in the substrate initially resulted in a decrease in nickel content, which was later followed by an increase in both aboveground and belowground components (Munsif et al., 2022). The incorporation of potassium silicate elevated aboveground heavy metals concentrations in sunflower seedlings, indicating the plant's capacity for nickel absorption (Dixit et al., 2022).

Previously, Shafiq et al. (2024) investigated the impact of nickel stress on flax plants revealed that nickel stress negatively impacts plant height, fresh biomass, and wheat yield due to reduced water absorption and nutrient deficiencies. Silicon-rich amendments in rice have shown a reduction in heavy metal accumulation and enhanced growth within multi-metal contaminated acidic soils (Maghsoudi et al., 2019). Nickel stress leads to the accumulation of reactive oxygen species (ROS), causing damage to nucleic acids, oxidation of proteins, and lipid peroxidation (Hafeez et al., 2024). The decrease in photosynthetic pigments due to nickel stress can be attributed to thylakoid membrane degradation, destruction of the chlorophyll apparatus, reduced chlorophyll synthesis, and decreased activity of protochlorophyll enzyme and chlorophyllase (Mahmoud and Fouad, 2024; Naveed et al., 2020). Recently, (Saleem et al., 2020) stated that treatment with silicon and copper, either individually or in combination, significantly improved plant growth, chlorophyll concentrations, and nucleic acid content in the shoots of flax genotypes. According to Rizwan et al. (2019), silicon significantly enhances plant growth and provides resistance to heavy metal toxicity in rice plants. It involves modifications to metabolic pathways, enhanced regulation of heavy metal uptake, increased antioxidant production, and the immobilization of toxic metal ions (Heile et al., 2021; Shah et al., 2024). Potassium silicate has the potential to enhance photosynthesis in wheat plants, likely linked to increased activity of photosynthetic enzymes such as ribulose-bisphosphate carboxylase, NADP+ dependent glyceraldehyde-3-phosphate dehydrogenase, and chlorophyll content under stress conditions (Liu and Liao, 2022; Moradi et al., 2024; Verma et al., 2021). The application of Si and Ni, individually or in combination, resulted in a significant enhancement of total phenolic, flavonoid, and α-tocopherol levels in the shoots of flax genotypes when compared to control plants (Shivappa et al., 2024). These findings provide light on the potential of silicate as an efficient strategy for strengthening the resistance of sunflowers to the effects of heavy metal stress. Subsequent research might investigate the optimum application rates, as well as the long-term effects on soil health and agricultural sustainability.

## CONCLUSION

Using potassium silicate under nickel stress raises the amount of chlorophyll, lowers the loss of electrolytes, boosts the activities of antioxidant enzymes, improves N-P-K uptake, makes it easier for potassium to be absorbed even when nickel is present, and ultimately leads to more growth and yield in sunflower crops. These findings highlight the possibility of silicate as an effective method for enhancing sunflower resilience to heavy metal stress. Subsequent research could investigate appropriate application rates and the long-term impacts on soil health and agricultural sustainability.

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