# Exogenous application of selenium on growth and antioxidant capacity of *Pisum sativum* L. under cadmium stress

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## Abstract

Heavy metals pose a serious threat to environment and show adverse effects on plants, animals, and human health. Cadmium is one of the highly persistent toxic metals present in agricultural fields due to its excessive release through industrial and anthropogenic activities. The present paper deals with the effect of sodium selenate on the growth and physiological parameters of *Pisum sativum* L. grown under cadmium stress. The application of sodium selenate significantly enhanced root and shoot length, biomass, and physiological attributes such as chlorophyll, sugar, proline, protein, and total antioxidant contents in pea plants, protecting them against cadmium toxicity. The highest total antioxidant content (54%) was reported in Cd (20 mM) + SS (5 µM) treatment. Hence, application of sodium selenate can be used as plant growth promoter for the growth of pea plants under cadmium stress.

**Keywords:** cadmium, growth, *Pisum sativum*, sodium selenate

**Kapoor, R. T., and M. Farzami Sepehr. 2023.** 'Exogenous application of selenium on growth and antioxidant capacity of *Pisum sativum* L. under cadmium stress'. *Iranian Journal of Plant Physiology 1*3 (1), 4389-4399**.**

## Introduction

The continuous application of wastewater for irrigation, excessive use of fertilizers, disposal of urban waste, smelting, increased industrialization, and anthropogenic activities have led to the enhanced level of heavy metal content and other pollutants in soil which affect quality and yield of crop plants worldwide (Gavrilescu, 2022). Most of the contaminants do not undergo any kind of chemical and microbial degradation and their concentration persists in soil for long duration (Kubier et al., 2019). The presence of metal in soil impairs balance in ecosystem, adversely affects human health via its entry in the food chain, reduces land for farming, and subsequently leads to food insecurity (Hassan et al., 2019; Zulfiqar et al., 2019; Hussain et al., 2021). Cadmium is recognized as a highly toxic metal which adversely affects health of animals and human beings (Chellaiah, 2018). Bojorquez et al. (2016) reported high concentration of cadmium in agricultural soils. The plants showed stunted growth when grown in cadmium contaminated soil (Bruno et al., 2017). Cadmium toxicity is known to induce oxidative stress by over-production of reactive

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*Received: June, 2022*
*Accepted: October , 2022*

oxygen species, damage cell membranes, biomolecules, and electron transport system, and interfere with nucleic acids and photosynthesis, resulting in reduced growth and development of plants (Abbas et al., 2017). Cadmium also induces osmotic stress in plants by minimizing relative water content, stomatal conductance and transpiration, thereby resulting in physiological damage in plants (Rizwan et al., 2016). The presence of cadmium in soil adversely affected morphological, physiological, and molecular characteristics of different plants (Qin et al., 2020; Haider et al., 2021). Hence, there is an urgent need to develop sustainable strategies to combat the unfavourable impacts of cadmium stress to maximize agricultural production.

Selenium is a metalloid belonging to the oxygen family in the periodic table and it has been recognized as an essential and beneficial element for plants. The average selenium content in soil has been reported approximately 0.1-2 mg/kg at global level (Chao et al., 2022). Selenium exists in inorganic and organic forms in nature. The inorganic forms are selenate, selenite, selenide, and elemental selenium, but the organic forms are selenocysteine and selenomethionine (Wu et al., 2015). Selenium and sulphur are the elements of chalcogens which show similar chemical properties. Selenite is found to be transported by phosphate transport mechanism whereas selenate is taken by sulfate transporters and metabolized along with the sulphur metabolic pathway in plants. Selenate is the most prevalent form of selenium in soil and it is more water soluble as compared to selenite (Missana et al., 2009). Selenium has been considered as double-edged sword as it is useful at low concentration but at higher concentration it reflects detrimental effects on plants and health of animals and human-beings (Naseem et al., 2021). At high concentrations, selenium replaces sulphur in amino acids and generates non-functional proteins and enzymes which contribute to its toxicity (Terry et al., 2000). Plants absorb inorganic selenium (selenite or selenate) from the soil and convert it into various organic selenides through sulphur metabolic pathway. Organoselenides promote plant growth, improve nutritional quality, and play an important role in physiological processes of plants (Chao et al., 2022). Selenium enhanced cellular functions like membrane stability, mineral nutrition, homeostasis, photosynthesis, antioxidant response and improved plant growth and development under metal/metalloid stress (Pandey and Gupta, 2015, Hasanuzzaman et al. 2022).

Pea (*Pisum sativum* L.; family: Fabaceae) also known as green pea or garden pea has been an important component of diet due to the presence of fiber, protein, starch, vitamins, and minerals like calcium, potassium, iron etc. (Kumari and Deka, 2021). Due to the less amount of saturated fat, cholesterol and sodium, it can be used by the patients of diabetes and cardiovascular diseases. *Pisum sativum* also shows antibacterial, antidiabetic, antifungal, and anti-inflammatory properties. As per our information, no reports are available about the mitigating role of sodium selenate against cadmium stress in pea plants. Considering the alleviating function of sodium selenate against metal toxicity, this investigation was planned to analyze the role of sodium selenate on growth and biochemical parameters of pea plants under cadmium stress.

## Material and Methods

### Plant material and chemicals

Healthy seeds of pea (*Pisum sativum* L. variety Vikas) were procured from seed agency of Ghaziabad, India. Seeds were stored in sterilized polythene bags to avoid contamination. Sodium selenate ($Na_2SeO_4$; 188.94 g/mol) was purchased from Merck. Analytical grade reagents were used for the experiments.

### Experimental design

Pea seeds were treated with 0.01% $HgCl_2$ solution for five minutes and washed with distilled water. Seeds of pea were kept for germination in the dark at 25±2 ℃ for seven days. The seedlings were cultivated in pots containing modified full-strength Hoagland's medium (Hoagland and Arnon, 1959). To study the effect of cadmium and selenium, the growth medium was supplemented with 20 mM $CdCl_2$ and 2.5 µM or 5 µM $Na_2SeO_4$ as per the treatment. The pots were kept at 25 ℃ (day/ night) with photoperiod 16 h/8 h (day/night) at photosynthetic active radiation 150 µmol $m^{-2}$ $s^{-1}$. During the experiment, the nutrient solution was continuously aerated and its losses were supplemented daily with distilled water. The medium was changed once in a week and its pH was kept at 5. The growth parameters of pea seedlings were measured after fifteen days.

**Seedling length and biomass estimation**

The root and shoot length of pea plants were measured with a measuring scale (ISTA, 2008). The fresh weight of pea seedlings was measured, then seedlings were kept in an oven at 65 ℃ for 72 hours and dry weight was estimated.

**Relative water content**

The fresh weight of pea seedlings was measured; then seedlings were immediately floated on distilled water at 25 ℃ under dark condition. After 12 h, turgid weight was taken and seedlings were dried in the oven at 80 ℃ for 48 h for the dry weight. Relative water content (RWC) was calculated by the modified method of Barrs and Weatherly (1962) using the following equation:

RWC (%) = (FW-DW) / (TW-DW) × 100

**Pigment content**

The pigment content was determined in pea leaves by the method of Lichtenthaler (1987). The pea leaves (10 mg) of control and treatment were ground with 10 ml of 80% acetone and centrifuged at 3000 rpm for 10 minutes. The optical density of the supernatant was measured at 645 and 663 nm and carotenoids content was determined at 470 nm:

Total chlorophyll (mg/g) = $20.2 \times OD_{645} + 8.02 \times OD_{663} \times V / 100 \times W$

Chlorophyll a (mg/g) = $12.7 \times OD_{663} - 2.69 \times OD_{645} \times V / 100 \times W$

Chlorophyll b (mg/g) = $22.9 \times OD_{645} - 4.68 \times OD_{663} \times V / 100 \times W$

where, V = volume of the supernatant in ml, W = fresh weight of leaves in g, and OD = optical density.

**Chlorophyll stability index (CSI)**

Chlorophyll stability index (CSI) was determined according to the method of Sairam et al. (1997) and calculated as below:

CSI = Total chlorophyll content in treatment/ Total chlorophyll content in control x 100

**Assessment of sugar content**

Total sugar content present in pea leaves was analyzed by the method of Hedge and Hofreiter (1962). Pea leaves (100 mg) were homogenized with 5 ml of 95% ethanol and centrifuged at 4000 g for 15 min. Supernatant (0.1 ml) was mixed with distilled water (0.9 ml) and 4 ml anthrone solution and the mixture was kept on water bath for 15 min. Absorbance was recorded at 620 nm after cooling and sugar content was calculated with reference to standard curve of glucose.

**Estimation of proline**

Proline content was analyzed by the method of Bates et al. (1973). Pea leaves were extracted with 3% sulphosalicylic acid and aliquot was treated with acid-ninhydrin and acetic acid and boiled for 1 h at 100 ℃. Reaction mixture was extracted with 4 ml toluene and the absorbance was measured at 520 nm. Proline content was expressed as µmol $g^{-1}$ FW using standard curve.

**Estimation of protein**

The protein content was measured by the method of Lowry et al. (1951). Pea leaves were homogenized with 1 N NaOH for 5 min at 100 ℃. Alkaline copper reagent was added to it and the mixture was kept at room temperature for 10 min, then Folin-Ciocalteu reagent was added. Absorbance of the solution was measured at 650 nm after 30 min and protein content was calculated with reference to BSA standard curve.

**Total antioxidant content**

The total antioxidant content in pea leaves was evaluated by Prieto et al. (1999). Total antioxidant capacity was analyzed in 0.1 ml sample solution (prepared by crushing 150 mg pea leaves in 3 ml ethanol) after mixing with 3 ml of reagent (0.6 M sulphuric acid, 28 mM sodium phosphate, and 4

Table 1
Effect of cadmium stress on root and shoot length, biomass, and relative water content of *Pisum sativum* L. var. Vikas with or without application of sodium selenate

| Treatment | Root length (cm) | Shoot length (cm) | Fresh weight (mg/g) | Dry weight (mg/g) | Relative water content (%) |
|---|---|---|---|---|---|
| Control | 7.19 ± 0.16 | 16.3 ± 0.14 | 6.44 ± 0.04 | 1.71 ± 0.07 | 88.48 ± 0.32 |
| Cd (20 mM) | 2.33 ± 0.18 | 4.14 ± 0.04 | 1.22 ± 0.01 | 0.31 ± 0.07 | 64.38 ± 0.10 |
| SS (5 µM) | 8.63 ± 0.15 | 19.32 ± 0.09 | 8.96 ± 0.22 | 2.44 ± 0.18 | 97.34 ± 0.04 |
| Cd (20 mM) + SS (2.5 µM) | 5.77 ± 0.14 | 8.58 ± 0.28 | 4.15 ±0.11 | 0.86 ± 0.01 | 78.73 ± 0.48 |
| Cd (20 mM) + SS (5 µM) | 6.81 ± 0.07 | 14.94 ± 0.51 | 5.38 ± 0.14 | 1.07 ± 0.08 | 83.57 ± 0.11 |

Data are means ± standard error of three independent experiments with three replicates. Whereas, Cd = Cadmium chloride and SS = Sodium selenate.

mM ammonium molybdate), and the absorbance was measured at 695 nm.

## Statistical Analysis

Experiments were conducted in triplicate and arranged in randomized block design. The data were determined using ANOVA and SPSS software (Version 16 SPSS, US). Mean data were assessed by DMRT at P< 0.05.

## Results

In the present investigation, the role of sodium selenate on growth and biochemical parameters of pea plants were assessed under cadmium stress.

### Growth of pea seedlings

Supplementation of sodium selenate showed a positive role by enhancing root and shoot length, relative water content, and biomass of pea seedlings under cadmium stress (Table 1). Maximum reductions 67.6% and 74.6% were observed in root and shoot length of pea plants under cadmium treatment, respectively over control. Significant enhancements by 39% and 42.7% respectively in fresh and dry weights of pea seedlings were reported under sodium selenate treatment as compared to control. The highest relative water content (97%) was observed in pea seedlings treated with sodium selenate (5 µM). The root and shoot lengths and biomass reflected the following trend: SS > C > $CdCl_2$ (20 mM) + SS (5 µM) > $CdCl_2$ (20 mM) + SS (2.5 µM) > $CdCl_2$.

### Pigment content

The pigment content in pea leaves showed significant increase with sodium selenate as compared to control and cadmium treatment. The significant increase in chlorophyll a, b, and total chlorophyll contents by 7.6%, 11.8%, and 9% were reported in pea leaves with sodium selenate treatment in comparison to control. Total chlorophyll and carotenoid contents showed the following trend: SS (5µM) > $CdCl_2$ (20 mM) + SS (5µM) > $CdCl_2$ (20 mM) + SS (2.5 µM) > $CdCl_2$ (20mM). The chlorophyll stability index (CSI) was highest in pea leaves with sodium selenate treatment, but maximum decrease was observed under cadmium stress (Fig. I.).

### Electrolyte leakage and lipid peroxidation

Cell membranes are considered as the first target site of free radical attacks generated under

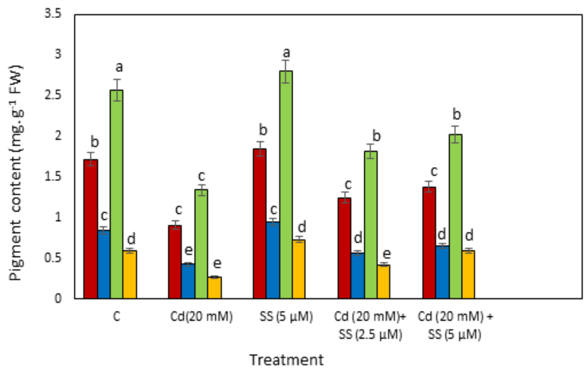


Fig. I. Effect of cadmium stress on pigment content of *Pisum sativum* L. var. Vikas with or without application of sodium selenate; dsata are mean ± standard error of three independent experiments with three replicates. Whereas, Cd = Cadmium chloride and SS = Sodium selenate.

Table 2
Effect of cadmium stress on sugar, proline, and protein contents of *Pisum sativum* L. var. Vikas with or without application of sodium selenate

| Treatment | Sugar content ($mg.g^{-1}$ DW) | Proline content ($\mu M.g^{-1}$ DW) | Protein content ($mg.g^{-1}$ FW) |
|---|---|---|---|
| Control | 3.15 ± 0.03 | 21.1 ± 0.31 | 11.86 ± 0.37 |
| $CdCl_2$ (20 mM) | 2.08 ± 0.07 | 35.26 ± 0.19 | 8.41 ± 0.12 |
| SS (5 µM) | 3.45 ± 0.13 | 43.28 ± 0.21 | 13.26 ± 0.18 |
| $CdCl_2$ (20 mM) + SS (2.5 µM) | 2.73 ± 0.08 | 41.11 ± 0.06 | 10.79 ± 0.11 |
| $CdCl_2$ (20 mM) + SS (5 µM) | 3.07 ± 0.04 | 46.28 ± 0.09 | 11.16 ± 0.01 |

Data are means ± standard error of three independent experiments with three replicates. Whereas, Cd = Cadmium chloride and SS = Sodium selenate.

cadmium stress. Free radicals also trigger oxidation of lipids, which stimulates increase in malondialdehyde (MDA) content, a by-product of the breakdown of membrane fatty acids and an indicator of the presence of free radicals (Pinto et al., 2016). Membrane damage was analyzed through electrolyte leakage and it was maximum (28.4%) in cadmium treated pea leaves (Fig. II). Electrolyte leakage and malondialdehyde content reflected the following order: $CdCl_2$ > $CdCl_2$ (20 mM) + SS (2.5 µM) > $CdCl_2$ (20 mM) + SS (5µM) > C > SS (5µM) (Fig. II). Results revealed that application of sodium selenate reduced electrolyte leakage and MDA content by 65.3% and 60.8%, respectively signifying its protective function against cadmium stress.

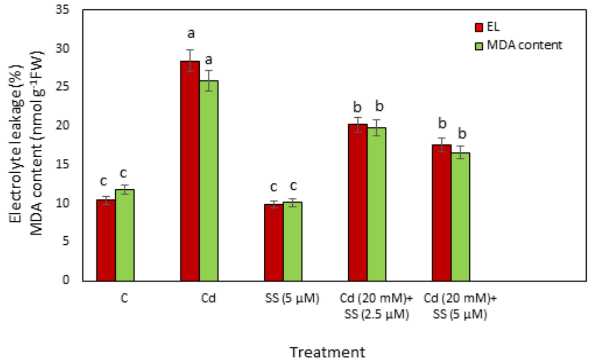


Fig. II. Effect of cadmium stress on electrolyte leakage and lipid peroxidation of *Pisum sativum* L. var. Vikas with or without application of sodium selenate; data are means ± standard error of three independent experiments with three replicates. Whereas, Cd = Cadmium chloride and SS = Sodium selenate.

### Physiological constituents

The physiological components such as sugar, proline and protein contents were also analyzed. Maximum sugar (3.45 mg/g) and protein (13.26 mg/g) contents were recorded in pea leaves with sodium selenate (5 µM) treatment (Table 2). Maximum increase in proline content (119%) was reported in pea leaves with sodium selenate as compared to control. Proline content showed the following trend: $CdCl_2$ (20 mM) + SS (5 µM) > SS > $CdCl_2$ (20 mM) + SS (2.5 µM) > $CdCl_2$ > Control.

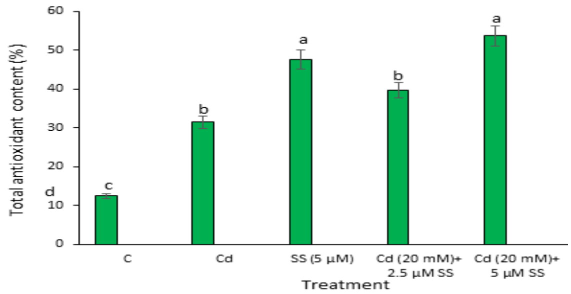


Fig. III. Effect of cadmium stress on total antioxidant content of *Pisum sativum* L. var. Vikas with or without application of sodium selenate

### Total antioxidant content

Total antioxidant content in *Pisum sativum* L. leaves was studied with or without sodium selenate supplementation and it reflected the following trend: $CdCl_2$ (20 mM) + SS (5 µM) > SS > $CdCl_2$ (20 mM) + SS (2.5 µM) > $CdCl_2$ (20 mM) > Control. Maximum total antioxidant content (53.6%) was observed in pea leaves treated with $CdCl_2$ (20 mM) + SS (5µM)

## Discussion

The beneficial effects of selenium have been reported on the growth of crop plants by earlier workers (Pandey and Gupta, 2015; Irmak, 2017). Selenium plays a crucial role in the protection of plants against the toxic effects of metals/metalloids (Feng et al., 2020; Riaz et al., 2021). The enrichment of soil with selenium alleviated the negative impacts of abiotic stresses such as salinity, UV-B radiation, drought, and toxicity of heavy metals in different plants (Haghighi et al., 2016). In the present study, selenium treatment promoted root and shoot growth and biomass and mitigated the adverse effects of cadmium on pea plants, possibly due to its less uptake or translocation of cadmium from roots to leaves (Table 1). There are reports that selenium inhibited uptake of toxic metal ions and their metabolism by rescuing their translocation from lower to upper plant parts which may be a vital metal/metalloid stress tolerance mechanism (Alyemeni et al., 2018; Gao et al., 2018; Hasanuzzaman et al., 2020). Alyemeni et al. (2018) reported less uptake of cadmium by tomato roots with reduced translocation into shoots and leaves under selenium treatment. Selenium supplementation enhanced the binding of metal ions to the root cell wall by improving pectin and hemicellulose composition and changed its distribution in roots as metal content decreased in plastids and mitochondria while it enhanced vacuolar sap and ribosomes (Zhao et al., 2019). Selenium at low concentration acts as an antioxidant stress alleviator and inhibits uptake of heavy metals such as mercury (Tran et al., 2018), chromium (Ulhassan et al., 2019), arsenic (Shahid et al., 2019) etc. in different plants. Feng et al. (2020) stated that selenium stimulated auxin production which assists in remodeling of root architecture for less metal uptake. The exogenous application of selenium reduced the concentration and influx of chromium by altering root morphology in *Brassica campestris* (Zhao et al., 2019). Ulhassan et al. (2019) observed reduction in chromium accumulation in *Brassica napus* with the application of selenium. The protective role of selenium improved phytochelatin or selenium - metal complex formation or detoxification of heavy metals and reduced metal transportation from roots to aerial plant parts for alleviation of metal toxicity (Hawrylak-Nowak et al., 2014). Handa et al. (2019) reported that application of selenium enhanced root and shoot length by 44% and 18%, respectively in *Brassica juncea* under chromium stress. Foliar application of selenium fertilizer enhanced the growth of peanut (Irmak, 2017).

Findings of the present study revealed that cadmium treatment reduced chlorophyll a, b, and total chlorophyll contents in pea leaves (Fig. I). The decrease in pigment content was mainly due to cadmium-induced reduction in photosynthesis, inhibition of electron transport chain, structural alterations in chloroplast, and stomatal aperture (Islam et al., 2008). Photosynthetic dysfunction due to the presence of heavy metals leads to the increased accumulation of ROS in plant cells and induced oxidative stress. Mozafariyan et al. (2014) observed that under cadmium stress, selenium enhanced fruit yield per plant through increasing photosynthetic pigments and improving antioxidant activities of pepper plants. Cadmium reduced uptake of iron and zinc by the plants resulting in chlorosis of leaves (Xu et al., 2017).

Supplementation of rice plants grown under lead and cadmium contaminated soil with nano-selenate improved plant growth and photosynthesis (Wang et al., 2021). Alves et al. (2020) reported that application of selenate resulted in increase in photosynthesis and biomass of tomato grown under cadmium stress. In wheat, combined application of selenium and zinc alleviated cadmium-induced reduction in growth, photosynthetic pigments, and photosynthesis (Wu et al., 2020). Foliar selenium treatment (50 g/ha) boosted chlorophyll synthesis and photosynthesis, enhancing antioxidant capacity as well as yield of cowpea plants (Silva et al., 2018).

Carotenoids are considered as antioxidants which can detoxify reactive oxygen species in plants and protect photosynthetic apparatus. Significant increase in carotenoid content was reported in pea leaves with sodium selenate; however, maximum reduction was observed with cadmium stress (Fig. I). Selenium application is one of the effective strategies to mitigate the toxic effects of metals as it improves biochemical functions of plants (Wang et al., 2020). In the present study, lipid peroxidation increased under cadmium stress which was reduced in the presence of sodium selenate. Proietti et al. (2013) also reported similar findings.

Sugar, proline, and protein act as osmoprotectant, play significant roles in maintenance of cell turgor, act as a driving force for absorption of water, and assist in scavenging of free radicals (Atteya and Amer, 2018). Proline acts as a compatible solute which accumulates under stress conditions and plays a pivotal role in osmoregulation (Annunziata et al., 2019). Proline protects plants by maintaining osmoregulation and detoxification of free radicals and preserves membrane integrity by stabilization of proteins and enzymes against environmental stresses. Table 2 shows that proline content was enhanced in the pea leaves in the presence of sodium selenate under cadmium stress. Proline prevents protein oxidation, reduces lipid peroxidation, and acts as a source of nitrogen and energy (Atteya et al., 2022).

Metal stress stimulates generation of reactive oxygen species, which causes the oxidative stress by damaging biomolecules such as lipids, protein, and DNA. The excessive production of free radicals under metal/metalloid can lead to plant death (Hasanuzzaman et al., 2020). Selenium induced improvement in cellular functions, membrane stability, and mineral nutrition with upregulation of antioxidants response, and reduced oxidative stress in plants as it has been widely reported against metal stress (Gupta and Gupta, 2017; Zhao et al., 2019; Wang et al., 2020). Figure (III) clearly depicts increase in total antioxidant activity in pea leaves in the presence of sodium selenate under cadmium stress, i.e. $CdCl_2$ (20 mM) + SS (5µM) treatment. Selenium application improved growth, photosynthesis, proline, and antioxidants level while reducing ROS production in quinoa (Khalofah et al., 2021). The presence of antioxidants boost plant defense system against oxidative stress. Selenium stimulated spontaneous dismutation of $H_2O_2$ and regulated antioxidant enzyme activities (Balakhnina and Nadezhkina, 2017).

Zhou et al. (2017) observed positive role of selenium in mitigation of mercury stress by enhancing antioxidative enzymes activities which promoted scavenging of ROS and reduced lipid peroxidation. Pandey and Gupta (2018) reported increase in the activity of several antioxidant enzymes such as catalase, superoxide dismutase, glutathione reductase, and peroxidase with an exogenous supply of selenium in rice plants under arsenic stress. Dai et al. (2019) observed positive role of selenium in *Brassica campestris* grown under zinc stress and proline content and antioxidant enzyme activities were enhanced with selenium addition. The foliar spraying of sodium selenate on wheat plants grown under cadmium-contaminated soil resulted in high biomass accumulation and antioxidant enzyme activity. Lipid peroxidation decreased whereas SOD, POD, CAT, and, APX increased significantly with the supplementation of selenium (Wu et al., 2020). Two different varieties of wheat (soft and durum) showed reduction in growth parameters under cadmium toxicity; however, supply of selenium via root and foliar routes reduced lipid peroxidation and increased peroxidase and superoxide dismutase activities in wheat plants (Zhou et al., 2021). In the present investigation, application of sodium selenate enhanced the growth of pea plants and reduced the negative impacts of cadmium stress. The utilization of sodium selenate is an environmentally benign, cost-effective, and sustainable approach for alleviation of cadmium toxicity in pea plants. Hence, sodium selenate can be used as a growth promoting fertilizer for plants under heavy metal toxicity.

## Conclusion

The findings of the present investigation revealed that cadmium stress inhibited growth of pea plants. It reduced chlorophyll, sugar, and protein contents whereas increased electrolyte leakage and lipid peroxidation in pea plants. Sodium selenate supplementation alleviated negative effects of cadmium stress in pea plants by triggering the up-regulation of carotenoids, proline, and total antioxidant content. Hence, sodium selenate can be used in agricultural fields for alleviation of heavy metal toxicity. Further in-depth investigations are required to explore its optimum concentration, exposure time, and mechanism of action as a fertilizer for growth and development of crop plants under heavy metal toxicity.

**Acknowledgements**

Dr. Riti Thapar Kapoor is thankful to Amity Institute of Biotechnology, Amity University Uttar Pradesh, Noida, India for offering laboratory facilities to carry out this study. .

## References

**Abbas, T., M. Rizwan, S. Ali, M. Adrees, M., Zia-ur-Rehman, M. F. Qayyum, Y.S. Ok, and G. Murtaza.** 2017. Effect of biochar on alleviation of cadmium toxicity in wheat (*Triticum aestivum* L.) grown on Cd-contaminated saline soil. *Environmental Science Pollution Research,* 25: 25668-25680.

**Annunziata, M.G., L.F. Ciarmiello, P. Woodrow, E. Dell' Aversana, and P. Carillo.** 2019. Spacial and temporal profile of glycine betaine accumulation in plants under abiotic stresses. *Frontiers in Plant Science,* 10: 230.

**Alves, L. R., D.R. Rossatto, M.L. Rossi, A.P. Martinelli, and P.L. Gratao.** 2020. Selenium improves photosynthesis and induces ultrastructural changes but does not alleviate cadmium-stress damages in tomato plants. *Protoplasma*, 257, 597–605

**Alyemeni, M.N., M.A. Ahanger, L. Wijaya, P. Alam, R. Bhardwaj, and P. Ahmad.** 2018. Selenium mitigates cadmium -induced oxidative stress in tomato (*Solanum lycopercicum* L.) plants by modulating chlorophyll fluorescence, osmolyte accumulation and antioxidant system. *Protoplasma,* 255, 985-986.

**Atteya, A.K.G. and H.M. Amer,** 2018. Influence of seaweed extract and amino acids on growth, productivity and chemical constituents of *Hibiscus sabdariffa* L. plants. *Bioscience Research,* 15: 772–791.

**Atteya, A.K.G., R.S. El-Serafy., K.M. El-Zabalawy., A. Elhakem, E.A.E. Genaidy.** 2022. Exogenously supplemented proline and phenylalanine improve growth, productivity, and oil composition of salted moringa by up-regulating osmoprotectants and stimulating antioxidant machinery. *Plants,* 11, 1553.

**Balakhnina, T. I., and E.S. Nadezhkina.** 2017. Effect of selenium on growth and antioxidant capacity of *Triticum aestivum* L. during development of lead-induced oxidative stress. *Russian Journal of Plant Physiology,* 64: 215–223

**Barrs, H.D., and P.E. Weatherley.** 1962. A re-examination of the relative turgidity technique for estimating water deficits in leaves. *Australian Journal* of *Biological Sciences,* 15:413-428.

**Bates, L.S., R.P. Waldren, and I.D. Teare.** 1973. Rapid determination of free proline for water-stress studies. *Plant Soil,* 39: 205-207.

**Bojorquez, C., M.G. Frias-Espericueta, D. Voltolina,** 2016. Removal of cadmium and lead by adapted strains of *Pseudomonas aeruginosa* and *Enterobacter cloacae*. *Revista Internacional de Contaminacion Ambiental,* 32: 407-412.

**Bruno,L., M. Pacenza, I. Forgione, L.R. Lamerton, M. Greco, A. Chiappetta, M.B. Bitonti.** 2017. In *Arabidopsis thaliana* cadmium impact on the growth of primary root by altering *SCR* expression and auxin-cytokinin cross-talk. *Frontiers in Plant Science,* 8: 1323.

**Chao,W., S. Rao, Q. Chen, W. Zhang, Y. Liao, J. Ye, S. Cheng, X. Yang, F. Xu,** 2022. Advances in research on the involvement of selenium in regulating plant ecosystems. *Plants,* 11: 2712.

**Chellaiah, E.R.** 2018. Cadmium (heavy metals) bioremediation by *Pseudomonas aeruginosa*: a mini-review. Appl. Water Sci. 8, 154.

**Dai, H., S. Wei, L. Skuza, and G. Jia.** 2019. Selenium spiked in soil promoted zinc accumulation of Chinese cabbage and improved its antioxidant system and lipid peroxidation. *Ecotoxicology and Environmental Safety,* 180: 179-184.

**Feng, R., L. Wang, J. Yang, P. Zhao, Y. Zhu, Y. Li, Y. Yu, H. Liu, C. Rensing, Z. Wu, R. Ni, and S. Zheng.** 2020. Underlying mechanisms responsible for restriction of uptake and translocation of heavy metals (metalloids) by selenium via root application in plants. *Journal of Hazardous Materials*, 402, 23570.

**Gao, M., J. Zhou, H. Liu, W. Zhang, Y. Hu, J. Liang, and J. Zhou.** 2018. Foliar spraying with silicon and selenium reduces cadmium uptake and mitigates cadmium toxicity in rice. *Science of the Total Environment,* 631: 1100-1108.

**Gavrilescu, M.** 2022. Enhancing phytoremediation of soils polluted with heavy metals. *Current Opinion in Biotechnology,* 74:21-31.

**Gupta,M., and Gupta, S.** 2017. An overview of seleniumuptake, metabolism, and toxicity in plants. *Frontiers in Plant Science,* 7:2074.

**Haghighi, M., A. Sheibanirad, and M. Pessarakli.** 2016. Effects of selenium as a beneficial element on growth and photosynthetic attributes of greenhouse cucumber. *Journal of Plant Nutrition,* 39(10): 1493–1498.

**Haider, F.U., L. Cai, J.A. Coulter, S.A. Cheema, J. Wu, R. Zhang, M. Wenjun, M. Farooq.** 2021. Cadmium toxicity in plants: Impacts and remediation strategies. *Ecotoxicology and Environmental Safety,* 211, 111887.

**Handa, N., S.K. Kohli, A. Sharma, A. K. Thukral, R. Bhardwaj, E.F. Abdallah, A. A. Alqarawi, P. Ahmad.** 2019. Selenium modulates dynamics of antioxidative defence expression, photosynthetic attributes and secondary metabolites to mitigate chromium toxicity in *Brassica juncea* L. plants. *Environmental and Experimental Botany,* 161: 180–192.

**Hasanuzzaman, M., M.B. Bhuyan, A. Raza, B. Hawrylak-Nowak, R. Matraszek- Gawron, J.A. Mahmud, K. Nahar, M. Fujita.** 2020. Selenium in plants: Boon or bane? *Environmental and Experimental Botany,* 178: 104170.

**Hasanuzzaman, M., K. Nahar, P. García-Caparrós, K. Parvin, F. Zulfiqar, N. Ahmed, and M. Fujita**. 2022. Selenium supplementation and crop plant tolerance to metal/metalloid toxicity. *Frontiers in Plant Science,* 12:792770.

**Hawrylak-Nowak, B., S. Dresler, and M. Wójcik.** 2014. Selenium affects physiological parameters and phytochelatins accumulation in cucumber (*Cucumis sativus* L.) plants grown under cadmium exposure. *Scientia Horticulturae,* 172:10–18.

**Hedge, J.E., and B.T. Hofreiter.** 1962. Estimation of carbohydrate. In Whistler, R.L.and Be Miller, J.N. (Ed.), Methods in carbohydrate chemistry. Academic Press, New York, pp. 17-22.

**Hoagland, D.R., and D.J. Arnon.** 1959. The water-culture method of growing plants without soil. *California Agricultural Experiment Station, Circular,* 347:26-29.

**Hussain, B., M.N. Ashraf, S.U. Rahman, A. Abbas, J. Lia, M. Farooq.** 2021. Cadmium stress in paddy fields: effects of soil conditions and remediation strategies. *Science of the Total Environment,* 754, 142188.

**Irmak, S.** 2017. Effects of selenium application on plant growth and some quality parameters in peanut (Arachis hypogaea). *Pakistan Journal of Biological Sciences,* 20(2):92-99.

**Islam, E., D. Liu, T.Q. Li, X. Yang, X. F. Jin, Q. Mahmooda, S. Tian, and J.Y. Li.** 2008. Effect of Pb toxicity on leaf growth, physiology and ultrastructure in the two ecotypes of *Elsholtzia argyi. Journal of Hazardous Materials,* 154 (1-3): 914-926.

**ISTA.** 2008. International rules for seed testing. international seed testing association. ISTA Secretariat, Switzerland.

**Khalofah, A., H. Migdadi, and E. El-Harty.** 2021. Antioxident enzymatic activities and growth response of quinoa (*Chenopodium quinoa* Willd.) to exogenous selenium application. *Plants,* 10, 719.

**Kubier, A., R.T. Wilkin, T. Pichler.** 2019. Cadmium in soils and groundwater: a review. *Applied Geochemistry,* 108, 104388.

**Kumari, T., and Deka, S.C.** 2021. Potential health benefits of garden pea seeds and pods: A review. *Legume Science,* 3:e82.

**Lichtenthaler, H.K.** 1987. Chlorophyll and carotenoids: pigments of photosynthetic biomembranes. In: Methods Enzymology. Packer L.,Douce R., (Eds.), Academic Press, Sandiego. pp. 350-382.

**Lowry, O.H., N.J. Rosebrough, A.L. Farr, and R.J. Randall,** 1951. Protein measurement with the Folin phenol reagent. *Journal of Biological Chemistry,* 193:265-275.

**Missana,T., U. Alonso, and M. García-Gutiérrez.** 2009. Experimental study and modelling of selenite sorption onto illite and smectite clays. *Journal of Colloid Interface Science,* 334: 132-138.

**Mozafariyan, M., L. Shekari, B. Hawrylak-Nowak, and M.M. Kamelmanesh.** 2014. Protective role of selenium on pepper exposed to cadmium stress during reproductive stage.

*Biological Trace Element Research,* 160: 97-107.

**Naseem, M., M. Anwar-ul-Haq, X. Wang, N. Farooq, M. Awais, H. Sattar, H. A. Malik, A. Mustafa, J. Ahmad, and M. A. El-Esawi**. 2021. Influence of selenium on growth, physiology, and antioxidant responses in maize varies in a dose-dependent manner. *Hindawi Journal of Food Quality,* 2021, Article ID 6642018, 1-9.

**Pandey, C., and M. Gupta.** 2015. Selenium and auxin mitigate arsenic stress in rice (*Oryza sativa* L.) by combining the role of stress indicators, modulators and genotoxicity assays. *Journal of Hazardous Materials,* 287: 384-391.

**Pandey, C., and M. Gupta.** 2018. Selenium amelioration of arsenic toxicity in rice shows genotypic variation: a transcriptomic and biochemical analysis. *Journal of Plant Physiology,* 231: 168-181.

**Pinto, S.S., A.E. Souza, M.A. Oliva, and E.G. Pereira.** 2016. Oxidative damage and photosynthetic impairment in tropical rice cultivars upon exposure to excess iron. *Scientia Agricola,* 73: 217-226.

**Prieto, P., M. Pineda, and M, Aguilar.** 1999. Spectrophotometric quantitation of antioxidant capacity through the formation of phosphomolybdenum complex: specific application to determination of vitamin E. *Analytical Biochemistry,* 269: 337-341.

**Proietti, P., L. Nasinia, D. del Buonoa, R. D'amatoa, E. Tedeschinib, and D. Businellia**. 2013. Selenium protects olive (*Olea europaea* L.) from drought stress. *Scientia Horticulturae,* 164: 165-171.

**Qin, S., H. Liu, Z. Nie, Z. Rengel, W. Gao, C. Li, P. Zhao.** 2020. Toxicity of cadmium and its competition with mineral nutrients for uptake by plants: a review. *Pedosphere,* 30:168-180.

**Riaz, M., M. Kamran, M. Rizwan, S. Ali, A. Parveen, Z. Malik, X. Wang.** 2021. Cadmium uptake and translocation: selenium and silicon roles in Cd detoxification for the production of low Cd crops: a critical review. *Chemosphere,* 273: 129690.

**Rizwan, M., S. Ali, T. Abbas, M.Z. Rehman, F. Hannan, C. Keller, M.I. Al-Wabel, Y.S. Ok.** 2016. Cadmium minimization in wheat: a critical review. *Ecotoxicology and Environmental Safety,* 130: 43-53.

**Sairam, R.K., P.S. Deshmukh, and D. S. Shukla.** 1997. Tolerance of drought and temperature stress in relation to increased antioxidant enzyme activity in wheat. *Journal of Agronomy and Crop Science,* 178: 171-178.

**Shahid, M.A., R.M. Balal, N. Khan, L. Zotarelli, G.D. Liu, A. Sarkhosh, J.C. Fernández-Zapata, J.J. Martínez Nicolás, and F. Garcia-Sanchez**. 2019. Selenium impedes cadmium and arsenic toxicity in potato by modulating carbohydrate and nitrogen metabolism. *Ecotoxicology and Environmental Safety,* 180: 588-599.

**Silva, V. M., E.H. M. Boleta, M.G. D.B. Lanza, J. Lavres, J. T. Martins, E.F. Santos, F.L.M. Santos, F.F. Putti, E. F. Junior, P.J. White, M.R. Broadley, H.W. P. de Carvalho, A.R. dosReis.** 2018. Physiological, biochemical, and ultrastructural characterization of selenium toxicity in cowpea plants. *Environmental and Experimental Botany,* 150: 172-182.

**Terry, N., A. M. Zayed, M.P. deSouza, and A.S. Tarun.** 2000. Selenium in greater plants. *Annual Review of Plant Physiology,* 51: 401-432.

**Tran, T. A. T., F. Zhou, W. Yang, M. Wang, Q.T. Dinh, D. Wang, D. Liang.** 2018. Detoxification of mercury in soil by selenite and related mechanisms. *Ecotoxicology and Environmental Safety,* 159: 77-84.

**Ulhassan, Z., R.A. Gill, H. Huang, S. Ali, T.M. Mwamba, B. Ali, Q. Huang, Y. Hamid, A.R. Khan, J. Wang, and W. Zhou**. 2019. Selenium mitigates the chromium toxicity in *Brassicca napus* L. by ameliorating nutrients uptake, amino acids metabolism and antioxidant defense system. *Plant Physiology and Biochemistry,* 145: 142-152.

**Wang, Q.Q., S.C. Yu, C.D. Xu, J.J. Liu, Y.Q. Li, M.H. Zhang, X.J. Long, Y.N. Liu, Y.F. Bi, W.H. Zhao, H.Y. Yao.** 2020. Association between selenium in soil and diabetes in chinese residents aged 35-74 years: results from the 2010 national survey of chronic diseases and behavioral risk factors surveillance. *Biomedical and Environmental Sciences*, 33: 260-268.

**Wang, C., T. Cheng, H. Liu, F. Zhou, J. Zhang, M. Zhang, X. Liu, W. Shi, T. Cao.** 2021. Nano selenium controlled cadmium accumulation and improved photosynthesis in indica rice cultivated in lead and cadmium combined paddy soils. *Journal of Environmental Sciences,* 103: 336–346.

**Wu, Z., G.S. Banuelos, Z.Q. Lin, Y. Liu, L. Yuan, X. Yin, M. Li.** 2015. Biofortification and phytoremediation of selenium in China. *Frontiers in Plant Science,* 6, 136.

**Wu, C., Y. Dun, Z. Zhang, M. Li, G. Wu.** 2020. Foliar application of selenium and zinc to alleviate wheat (*Triticum aestivum* L.) cadmium toxicity and uptake from cadmium-contaminated soil. *Ecotoxicology and Environmental Safety,* 190: 110091.

**Xu, Z.M., Q.S. Li, P. Yang, H.J. Ye, Z.S. Chen, S.H. Guo, L.L. Wang, B.Y. He, E. Y. Zeng.** 2017. Impact of osmoregulation on the differences in Cd accumulation between two contrasting edible amaranth cultivars grown on Cd-polluted saline soils. *Environmental Pollution,* 224: 89-97.

**Zhao, Y., C. Hu, X. Wang, X. Qing, P. Wang, Y. Zhang, X. Zhang, X. Zhao.** 2019. Selenium alleviated chromium stress in Chinese cabbage (*Brassica campestris* L. sp. Pekinensis) by regulating root morphology and metal element uptake. *Ecotoxicology and Environmental Safety,* 173: 314-321

**Zhou, J., Z. Jiang, J. Ma, L. Yang, and Y. Wei.** 2017. The effects of lead stress on photosynthetic function and chloroplast ultrastructure of *Robinia pseudoacacia* seedlings. *Environmental Science and Pollution Research,* 24: 10718-10726.

**Zhou, J., C. Zhang, B. Du, H. Cui, X. Fan, D. Zhou, J. Zhou.** 2021. Soil and foliar applications of silicon and selenium effects on cadmium accumulation and plant growth by modulation of antioxidant system and Cd translocation: comparison of soft vs. durum wheat varieties. *Journal of Hazardous Materials,* 402, 123546.

**Zulfiqar, U., M. Farooq, S. Hussain, M. Maqsood, M. Hussain, M. Ishfaq, M. Ahmad, M. Z. Anjum.** 2019. Lead toxicity in plants: impacts and remediation. *Journal of Environmental Management,* 250, 109557.