



Research Article

Morphological Responses of Wheat (*Triticum aestivum*) and Mustard (*Brassica nigra*) to Plant-Derived Smoke Solution Under Heavy Metal Salt Stress

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Abstract | Heavy metal stress, particularly from cadmium (Cd), represents a major abiotic constraint that significantly reduces crop productivity. Plant-derived smoke (PDS) has previously been reported to enhance seed germination, seedling growth, and biomass accumulation in crops exposed to heavy metal stress. In line with these findings, the present study was designed to evaluate the effect of PDS solution on the morphological traits of wheat and mustard under cadmium sulfate (CdSO₄) stress. A completely randomized design (CRD) with five replications per crop was employed using wheat and black mustard. Smoke solution was prepared from *Cymbopogon jwarancusa*. Nine treatment combinations consisting of CdSO₄ alone (30, 60, 90 mM), PDS alone (10, 50, 100 ppm), and their mixtures (30+1000, 60+1000, 90+1000 ppm Cd+PDS) were applied to seeds, with four replicates each. Data were recorded for germination percentage, plumule length (cm), radical length (cm), fresh weight (g), dry weight (g), and moisture content (%). The results demonstrated that PDS applied either alone or in combination substantially enhanced plant growth attributes, counteracting Cd toxicity and confirming its promising mitigating role. Statistical accuracy was verified using the LSD test at alpha levels of 0.05, 0.01, and 0.001. While Cd alone reduced growth rates, PDS significantly promoted growth compared to the control. It is concluded that PDS could serve as a strong candidate for alleviating the negative effects of heavy metals on plant growth in contaminated soils.

Received | January 16, 2025; **Accepted** | June 06, 2026; **Published** | June 19, 2026

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Citation | Nazish, S. Naveed, Kalsoom, Riffat and I.U. Khan. 2026. Morphological responses of wheat (*Triticum aestivum*) and mustard (*Brassica nigra*) to plant-derived smoke solution under heavy metal salt stress. *Pakistan Journal of Weed Science Research*, 32(2): 144-148.

DOI | <https://dx.doi.org/10.17582/journal.pjwsr/2026/32.2.144.148>

Keywords | Cadmium, Heavy metal stress, Mustard, Smoke solution, Wheat



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Introduction

Abiotic stresses including extreme temperatures, drought, salinity, and heavy metals are major contributors to global pollution. Heavy metals,

originating from both natural and anthropogenic sources, are released through various activities. Although some metals (e.g., copper, zinc, nickel, iron) are essential for living organisms, others such as arsenic, cadmium, chromium, and mercury are non-

essential and exert toxic effects (Bibi *et al.*, 2023). Cadmium, a highly phytotoxic heavy metal with high solubility in soil, impairs plant growth and reduces yield by interfering with plant physiological systems.

Wheat (*Triticum aestivum*; family *Poaceae*) is one of the most widely cultivated cereal crops and a staple food in Pakistan and worldwide. Mustard (*Brassica nigra*), a major edible oil source, is often grown alongside or in rotation with wheat during winter. Both crops have been reported to be adversely affected by heavy metal stresses, including Cd (Mourato *et al.*, 2015).

Plant-derived smoke, a complex mixture of biologically active compounds, has been shown to enhance plant growth. Decades ago, PDS was used to promote seed germination in endangered plant species (De Lange and Boucher, 1990). Active compounds such as karrikins and cyanohydrin (Khatoon *et al.*, 2020) break seed dormancy and improve plant growth (Van *et al.*, 2000). These compounds exhibit high heat stability in solution and remain effective at very low concentrations (Flematti *et al.*, 2004). Moreover, PDS treatments have been reported to improve plant growth and vigor under various stress conditions, including heavy metal, drought, salinity, and temperature stresses (Ibrahim *et al.*, 2022).

The objective of this study was to test whether different concentrations of PDS could mitigate cadmium sulfate stress in wheat and mustard seeds.

Materials and Methods

Seeds of wheat (*Triticum aestivum*) and mustard (*Brassica nigra*) were obtained from the agriculture center of Karak and sterilized with 50% ethanol for 5 minutes, followed by three rinses with distilled water.

A standard petri dish bioassay was used for germination studies. Seeds were placed on filter paper in petri dishes, and each treatment was replicated four times. Dishes were arranged randomly and maintained under controlled laboratory conditions.

Treatments

Seeds were treated with the following nine solutions:

1. Control: Tap water.
2. Cadmium stress: Three CdSO_4 concentrations (30, 60, and 90 mM).
3. Smoke water: PDS solutions at 10, 50, and 100

ppm.

4. Combined treatments: Each CdSO_4 concentration (30, 60, 90 mM) mixed with 1000 ppm smoke solution.

Preparation of smoke solution

Smoke solution was prepared by burning semi-dried *Cymbopogon jwarancusa* plant material in a sealed furnace. The smoke was bubbled through chilled distilled water to produce a concentrated stock solution (1000 ppm), which was then filtered and diluted as needed.

Statistical analysis

Results were analyzed using one-way analysis of variance (ANOVA). Differences between treatment means were tested for significance using the Least Significant Difference (LSD) test at $\alpha = 0.05$, 0.01, and 0.001.

Results

Seed germination

Cadmium stress markedly suppressed seed germination in both wheat and mustard. Germination declined from 98% in the control to only 36% at the highest Cd concentration (90 mM). PDS treatments alone improved germination, achieving rates between 94% and 100%. Most notably, when PDS was applied together with cadmium, it largely reversed the inhibitory effect, raising wheat germination from 36% to 80%.

Shoot and root length

Cadmium treatment negatively affected seedling growth, causing stunted development. Wheat shoot length was reduced to less than 0.2 cm, and mustard shoots failed to develop at all. Root growth was similarly shortened. PDS alone significantly promoted shoot and root development. In combined treatments, PDS alleviated Cd-induced damage, resulting in significantly longer shoots and roots compared to Cd-only treatments.

Biomass

Cadmium stress caused a major reduction in seedling fresh and dry weight. PDS application alone increased biomass relative to the control. In combined treatments, seedlings exhibited significantly greater biomass than those under Cd stress alone, indicating that PDS partially counteracted metal toxicity.

Discussion

Seed germination is a critical stage determining crop production success. In this study, germination of wheat and mustard was observed under various Cd concentrations (30, 60, 90 mM), PDS concentrations (10, 50, 90 ppm), and their combinations. [Vijayarengan \(2012\)](#) reported that increasing Cd concentrations inhibited seedling vigor and plant biomass. Similarly, [Rosca et al. \(2021\)](#) found that seed germination and seedling growth decrease with increasing metal doses. Our results align with these reports: Cd reduced germination rates, radical/plumule length, fresh weight, dry weight, and moisture content, whereas PDS accelerated germination and plant development.

[Mihalescu et al. \(2010\)](#) noted that reduced root and shoot height resulted from Cd accumulation, with the reduction being directly proportional to metal concentration. Several studies ([Correa et al., 2006](#); [Ahmad et al., 2008](#); [Jun-yu et al., 2008](#); [Ahmad et al., 2011](#)) confirmed that Cd has a strong inhibitory effect on seed germination and morphological parameters, particularly root and shoot length, due to high Cd toxicity. [Mukhtar et al. \(2012\)](#) observed decreased fresh and dry weight of two native grasses with increasing Ni and Cd levels. [Pirselova \(2021\)](#) also reported reductions in fresh weight, dry weight, and water content under applied Cd doses. Our results showed a gradual decrease in fresh and dry weight of both crops with increasing metal concentration.

[Jamil et al. \(2014\)](#) reported that smoke-treated seeds exhibited better germination percentages than water-treated controls. According to [Daws et al. \(2007\)](#), PDS produces hormone-like responses in various species, interacting with auxins, gibberellins, and cytokinins. Current results demonstrated that PDS improved root and shoot length, with lower dilutions promoting better germination than higher dilutions. [Sparg et al. \(2006\)](#) explained that lower smoke dilutions yield better germination because important bioactive chemicals become diluted at higher concentrations, reducing their positive effects. [Iqbal et al. \(2016\)](#) found that PDS positively affects seed germination and increases leaf area, root length, shoot length, and plant biomass. [Ibrahim et al. \(2022\)](#) reported that while As and Hg reduced wheat seedling growth, PDS increased seedling growth both individually and in combination with these metals. [Hayat et al. \(2022\)](#) similarly observed that PDS alone and PDS+salt

solutions enhanced seed germination. Our findings are consistent with these studies, showing that PDS restores root and shoot length, fresh and dry weight, and moisture content under salt stress.

Conclusion

The present findings indicate that heavy metal stress (cadmium sulfate) significantly decreased morphological parameters in wheat and mustard. Conversely, *Cymbopogon jwarancusa* smoke solution enhanced seed germination and increased plant biomass. The combination of PDS with CdSO₄ alleviated the inhibitory effects of CdSO₄ on wheat and mustard seedlings.

Future Recommendations

1. Isolation of biologically active compounds from smoke responsible for its mitigating effects is recommended.
2. Research should focus on understanding the beneficial effects of smoke at the gene expression level.
3. Smoke water should be tested practically as a seed primer under real field conditions using polluted soils.

Acknowledgement

The authors are thankful to the Lab supervisory staff for help and support.

Novelty Statement

This research innovatively establishes the efficacy of *Cymbopogon jwarancusa* derived smoke solution as a eco-friendly treatment to counteract cadmium-induced stress and promote growth in wheat and brassica, presenting a viable approach for agricultural remediation.

Author's Contribution

Shahida Naveed, Nazish, Kalsoom and Riffat designed, conducted and analyzed data and Inayat Ullah Khan provided technical and financial support.

Generative AI and AI assisted technology statement

The authors declare that no generative AI and AI assisted technology was used in the creation of this manuscript.

Conflict of interest

The authors have declared no conflict of interest.

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