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ASSESSMENT OF LEAD, CADMIUM, IRON, AND ZINC CONTAMINATION IN WHEAT GRAINS FROM MULTAN DISTRICT: SOURCES, HEALTH RISK EVALUATION, AND POTENTIAL IMPLICATIONS

Original Research

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ABSTRACT

Background: Wheat (*Triticum* spp.) is a staple food crop cultivated globally and consumed by over one-third of the world's population. In Pakistan, it remains a primary dietary source of macronutrients and essential trace minerals such as iron and zinc. However, the rise of heavy metal contamination—mainly from industrial runoff, wastewater irrigation, vehicular emissions, and agrochemicals—has raised serious concerns about food safety, particularly with respect to cadmium (Cd), lead (Pb), iron (Fe), and zinc (Zn).

Objective: This study aimed to quantify the concentration of selected heavy metals in commonly grown wheat varieties from Multan district, Pakistan, and assess potential public health risks associated with their consumption.

Methods: A total of 25 wheat grain samples representing five different varieties (A–E) were collected from local farms, markets, and storage facilities. Parallel soil and irrigation water samples were obtained from corresponding fields. All samples were subjected to wet acid digestion and analyzed using Flame Atomic Absorption Spectrophotometry (FAAS). Statistical comparisons were conducted using ANOVA and homogeneous grouping to evaluate inter-varietal metal differences.

Results: Lead (Pb) concentration ranged from 0.0320 ± 0.0261 to 0.0376 ± 0.0355 mg/kg, with significant variation across varieties (p < 0.05). Zinc (Zn) ranged from 0.0557 ± 0.0283 to 0.1153 ± 0.0546 mg/kg and also showed significant varietal differences. Cadmium (Cd) ranged between 0.0338 ± 0.0070 and 0.0391 ± 0.0060 mg/kg, and iron (Fe) varied from 0.0396 ± 0.0052 to 0.0850 ± 0.0041 mg/kg; both showed statistically non-significant differences (p > 0.05). All concentrations remained below international safety thresholds but indicated potential cumulative exposure risk.

Conclusion: The study highlights low but detectable levels of heavy metals in wheat grains from Multan, especially Pb and Zn. It recommends proactive measures such as using clean irrigation sources, soil monitoring, phytoremediation practices, and safe grain storage to reduce contamination. These findings support the promotion of sustainable agricultural practices and informed policymaking to protect food safety and public health.

Keywords: Cadmium, Flame Atomic Absorption Spectrophotometry, Food Safety, Iron, Lead, Multan, Zinc.

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INTRODUCTION

Wheat (*Triticum* spp.) remains one of the most significant cereal crops globally, cultivated across diverse agroecological zones and serving as a staple food source for nearly two-thirds of the world's population. It is not only an energy-dense food but also provides essential nutrients such as proteins, dietary fiber, B-complex vitamins, iron, zinc, and magnesium, making it a vital component in addressing global nutritional needs (1–4). In Pakistan, wheat is cultivated on approximately 8 million hectares with an annual per capita consumption of around 120 kg, signifying its central role in the national diet and food security system (3,4). Over the past 8000 years, wheat has evolved from ancient, low-yielding but resilient cultivars to modern, high-yielding varieties responsive to fertilizers, marking a significant transformation during and after the Green Revolution (2,5). While wheat's nutritional contributions are undeniable, its safety has come under increasing scrutiny due to the risk of heavy metal contamination. Cadmium (Cd), lead (Pb), iron (Fe), and zinc (Zn) are among the common heavy metals that may accumulate in wheat grains, especially when crops are irrigated with untreated or poorly treated wastewater (6,7). These metals can enter agricultural systems through various anthropogenic activities such as industrial emissions, mining, the use of phosphate-based fertilizers and pesticides, as well as from recycled materials and traffic emissions near urban and roadside fields (8). In some cases, even natural weathering of metal-rich rocks can contribute, albeit to a lesser extent (9). The continued use of contaminated irrigation water not only degrades soil health but also leads to bioaccumulation of toxic elements in wheat, thereby compromising food safety and human health (10).

Toxicological concerns associated with heavy metals are well documented. Prolonged cadmium exposure is known to impair renal function and respiratory health and is associated with bone demineralization, while lead adversely affects neurological development and cardiovascular health (11-13). Although iron and zinc are essential trace elements, their excess—especially in the form of bioavailable free ions—can cause oxidative stress, cellular damage, and neurological effects (14). In addition, heavy metal interference may reduce the bioavailability of beneficial nutrients like zinc and iron in wheat, thereby exacerbating malnutrition and increasing the risk of chronic diseases in human populations reliant on wheat as a dietary staple (15,16). Moreover, such contamination undermines the agricultural value of wheat and compromises consumer confidence in food safety (17). In regions like South Asia, particularly Pakistan, the intersection of wastewater irrigation, industrial expansion, and limited regulatory oversight presents a critical public health challenge. While previous studies have addressed the presence of heavy metals in crops, there remains a gap in understanding varietal differences in metal uptake and the associated health risks. This warrants a systematic evaluation of commonly grown wheat cultivars in contaminated environments to determine the extent of metal accumulation and its implications for human consumption (17,18). In light of these concerns, the present study was undertaken to assess the uptake of cadmium, lead, iron, and zinc in selected wheat varieties cultivated in the Multan region of Pakistan, where wastewater irrigation is prevalent. It also aims to evaluate the associated health risk for consumers, thereby providing a scientific basis for recommending safer agricultural practices and potential breeding strategies for low metal-accumulating wheat varieties (19).

METHODS

The present study was conducted over a one-year period, from May 2023 to April 2024, across multiple agricultural zones within the Multan district of Pakistan. It aimed to evaluate the presence and concentration of selected heavy metals—cadmium (Cd), lead (Pb), iron (Fe), and zinc (Zn)—in wheat (*Triticum* spp.), soil, and irrigation water to assess environmental exposure and potential health risks. A cross-sectional sampling strategy was adopted to obtain representative samples from cultivated fields, grain storage facilities, and irrigation sources. Ethical approval for this study was obtained from the Institutional Review Board of Bahauddin Zakariya University, Multan and all necessary permissions from farm owners and market vendors were secured prior to sampling. Wheat samples were collected using aseptic techniques to avoid contamination during handling. Approximately 250 grams of wheat grains were gathered from local farms, storage units, and grain markets using sterile gloves and sealed in sterile, labeled containers (1–3). These were promptly transported to the Food Safety and Microbiological Laboratory at Bahauddin Zakariya University for further processing. Parallel to wheat sampling, soil samples were collected from the same cultivated fields. Surface soil from a depth of 0–15 cm around the root zones was collected using clean tools and protective gloves to avoid cross-contamination. Each 200-gram sample was packed into labeled zip-



lock bags and transported at ambient temperature to the laboratory (4–7). Irrigation water samples were also obtained from these fields, collected in sterile glass bottles, carefully labeled, and refrigerated until laboratory transfer (8–10).

All wheat, soil, and water samples underwent wet digestion for heavy metal quantification. Standard digestion protocols were followed, utilizing concentrated nitric acid (60%) and perchloric acid (70%) to break down the organic matrix and release metal ions for analysis (11–13). For the wheat grain samples, approximately 1 gram of finely ground powder was weighed and placed into digestion flasks. Ten milliliters of nitric acid were added, and the flasks were sealed with aluminum foil and left in the dark for 24 hours (15–17). Subsequently, 5 milliliters of perchloric acid were added, and the mixture was allowed to stand at room temperature for four hours before being heated on a hot plate at 200°C until a clear solution was achieved (18,19). The same protocol was applied to soil samples after air drying and sieving (9,10). For irrigation water, a 1 mL aliquot was digested using identical acid mixtures and heating conditions (11,12). Post digestion, all samples were allowed to cool and then diluted with double-distilled water to a final volume of 50 mL (12,13). Filtration was performed using Whatman No. 42 filter paper, and the filtrates were collected in clean, labeled glass containers for subsequent analysis (12-14). The concentrations of Cd, Pb, Zn, and Fe in all sample matrices were measured using Atomic Absorption Spectrophotometry (AAS), a reliable and sensitive analytical technique for trace metal detection (15,16). Calibration curves were prepared using certified standard solutions to ensure measurement accuracy. Each sample was analyzed individually, and results were digitally recorded and subjected to statistical interpretation using appropriate software tools (14–16).

RESULTS

The quantitative analysis of wheat varieties collected from the Multan region revealed varying levels of heavy metal concentrations, specifically cadmium (Cd), lead (Pb), iron (Fe), and zinc (Zn). All values recorded for heavy metals in wheat grains were within permissible limits for human consumption, though varietal differences were observed in lead and zinc concentrations. The data also provided insight into the distribution of these metals, emphasizing areas of significant and non-significant variation across the cultivars.

Concentration of Lead (Pb) in Wheat Samples: Lead concentrations demonstrated significant inter-varietal differences (p < 0.05), resulting in two distinct homogeneous groups. The highest Pb content was detected in Variety A (0.0376 \pm 0.0355 mg/kg), followed by Variety B (0.0357 \pm 0.0391 mg/kg) and Variety C (0.0354 \pm 0.0541 mg/kg), both sharing characteristics of Groups A and B. Varieties D and E exhibited the lowest Pb levels, with concentrations of 0.0322 \pm 0.0135 mg/kg and 0.0320 \pm 0.0261 mg/kg respectively. Despite the statistical significance, the absolute differences in Pb content were minimal, and all values remained well within permissible exposure thresholds, implying a low health risk under typical consumption levels (17-19).

Concentration of Cadmium (Cd) in Wheat Samples: Cadmium concentrations showed no statistically significant variation among the tested wheat varieties (p > 0.05), with all samples falling within a single homogeneous group. Variety D recorded the highest cadmium level at 0.0391 ± 0.0060 mg/kg, followed by Variety C (0.0384 ± 0.0061 mg/kg) and Variety E (0.0381 ± 0.0086 mg/kg). The lowest values were found in Variety B (0.0338 ± 0.0070 mg/kg), with Variety A slightly higher at 0.0345 ± 0.0070 mg/kg. These findings suggest uniform environmental exposure and uptake patterns of Cd across the selected varieties (20-22).

Concentration of Iron (Fe) in Wheat Samples: The iron content in wheat grains did not differ significantly between varieties (p > 0.05), with all samples categorized into a single group. Variety C had the highest iron level at 0.0850 ± 0.0041 mg/kg, whereas Variety D recorded the lowest concentration at 0.0396 ± 0.0052 mg/kg. Varieties A, B, and E fell in between, with iron levels of 0.0484 ± 0.0015 mg/kg, 0.0669 ± 0.0022 mg/kg, and 0.0412 ± 0.0035 mg/kg respectively. These relatively stable iron values indicate minimal influence of varietal genetics or field conditions on Fe uptake (23,24).

Concentration of Zinc (Zn) in Wheat Samples: Zinc concentrations differed significantly among the varieties (p < 0.05), forming two homogeneous groups. Variety B (0.1153 \pm 0.0546 mg/kg) and Variety C (0.1087 \pm 0.0534 mg/kg) had the highest levels, while Variety A showed an intermediate value of 0.0869 \pm 0.0063 mg/kg. Varieties D and E demonstrated the lowest Zn concentrations, recorded at 0.0569 \pm 0.0102 mg/kg and 0.0557 \pm 0.0283 mg/kg, respectively. These differences aligned with regional disparities reported in other studies and may be influenced by local soil composition and agricultural practices (25,26).



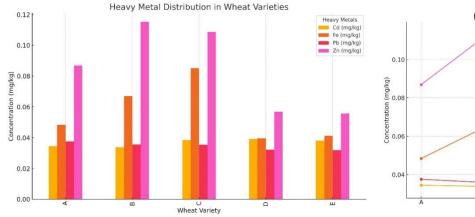
Table 1: Specific Sources of Heavy metals found in Wheat

Metals	Source	Reference
Lead	Polyvinyl chloride (PVC) pipes used in sanitation, agricultural tools, recycled PVC lead,	(10,11)
	batteries, lunch boxes, jewelry, etc.	
Cadmium	pigments, photovoltaic cells, electroplated components, batteries, paints, polymers,	(12)
	photoconductors, synthetic rubber, and other materials used in photography and engraving.	
Iron	Industrial emission, mining activities, iron-rich fertilizers, pesticides, domestic-agricultural	(13,15)
	waste and equipment contamination etc.	
Zinc	mining, smelting, industrial processes, post-harvest handling and storage in zinc coated	(14)
	containers etc.	

Table 2: Showing Results for Cd, Fe, Pb, and Zn by Wheat Grains

Variety	Cd (mg/kg)	Fe (mg/kg)	Pb (mg/kg)	Zn (mg/kg)
A	0.0345 ± 0.006975^a	$0.0484 \pm 0.001503~^{\rm a}$	$0.0376 \pm 0.035535 ^{\rm a}$	$0.0869 \pm 0.006274^{\rm \ a}$
В	$0.0338 \pm 0.006999~^{\rm a}$	$0.0669 \pm 0.002202~^{\rm a}$	$0.0357 \pm 0.03909 ~^{ab}$	0.1153 ± 0.05461 a
С	$0.0384 \pm 0.006107^{\ a}$	$0.0850 \pm 0.004137^{\rm \ a}$	$0.0354 \pm 0.054075 ^{ab}$	$0.1087 \pm 0.053393 \text{ ab}$
D	$0.0391 \pm 0.005954^{\rm \ a}$	$0.0396 \pm 0.005157^{\mathrm{a}}$	0.0322 ± 0.013545^{b}	0.0569 ± 0.01018 b
Е	$0.0381 \pm 0.008643~^{\rm a}$	$0.0412 \pm 0.003534^{\rm \ a}$	0.0320 ± 0.026122^{b}	0.557 .028275 b

Note: Homogeneous groups for Cd and Fe indicate no significant differences among varieties (p > 0.05). For Pb and Zn, letters (a, ab, b) indicate significant groupings (p < 0.05). Data are presented as mean \pm standard deviation.



Heavy Metal Concentrations in Wheat Varieties

O.10

O.04

A

B

Cd (mg/kg)

Fe (mg/kg)

Fe (mg/kg)

Zn (mg/kg)

D

E

Wheat Variety

Figure 1 Heavy Mental Distribution in Wheat Varieties

Figure 2 Heavy Mental Concentrations in Wheat Varieties

DISCUSSION

The findings of this study provide a comprehensive analysis of the heavy metal accumulation in commonly cultivated wheat varieties in the Multan region of Pakistan, with a focus on cadmium (Cd), lead (Pb), iron (Fe), and zinc (Zn). The results revealed that while Cd and Fe concentrations showed no statistically significant inter-varietal differences, Pb and Zn did exhibit slight but meaningful variations. These findings align with prior regional assessments, which have reported that wheat grown in areas exposed to industrial or untreated wastewater irrigation tends to accumulate higher levels of toxic elements, particularly Pb and Cd, posing significant public health risks if not regularly monitored (18-20). The lack of significant variation in cadmium and iron content across the varieties suggests a uniform exposure to contamination sources, possibly through irrigation practices, atmospheric deposition, or consistent agrochemical usage. The slightly higher Cd levels in some varieties, though not statistically significant, remain a concern due to Cd's cumulative toxicity and its known nephrotoxic and osteotoxic effects in humans (21). Likewise, the homogeneity in Fe levels indicates minimal



varietal influence on Fe uptake under the prevailing soil and environmental conditions, though bioavailability of Fe could be compromised if it interacts antagonistically with Cd or Pb in the rhizosphere (22).

In contrast, the significant inter-varietal differences observed in Pb and Zn concentrations highlight the influence of genetic traits and micro-environmental variables. Pb is particularly concerning due to its neurotoxic potential, especially in vulnerable populations such as children and pregnant women. Although the measured concentrations remained below internationally accepted thresholds, even low-level chronic exposure can have long-term health consequences (23). The relatively higher Zn levels in certain varieties were within the optimal range and are beneficial for human health, as Zn plays a critical role in immune function and enzyme activity. However, imbalanced Zn levels can also interfere with iron absorption and vice versa, necessitating a holistic interpretation of nutrient-metal interactions (24). These findings underscore the dual nutritional and toxicological role of trace elements in cereal crops. The presence of essential metals like Fe and Zn, while nutritionally valuable, can be compromised by the concurrent uptake of toxic elements. Hence, monitoring these metals is critical not only from a food safety standpoint but also from a nutritional bioavailability perspective. Moreover, the wheat varieties in this study displayed Pb and Cd levels consistent with reports from similar agroecological settings in India and Bangladesh, reinforcing the transboundary nature of environmental contamination in South Asia (25,26).

One of the strengths of this study lies in its comprehensive sampling design, encompassing field, market, and storage-level grain assessment, thereby reflecting real-world exposure scenarios. The use of atomic absorption spectrophotometry (AAS) also lends analytical rigor and sensitivity to the heavy metal quantification. However, there are limitations that must be acknowledged. The study did not assess the corresponding heavy metal content in human biological samples, such as hair or blood, which would strengthen the linkage between environmental exposure and health outcomes. Furthermore, no soil physico-chemical data were included, which could help explain metal mobility and bioavailability. Seasonal variation was also not accounted for, although fluctuations in metal concentrations due to climatic changes or irrigation cycles can influence uptake dynamics. Given the accumulating evidence on heavy metal risks in agricultural systems, these findings advocate for an integrated risk management approach. This includes promoting the use of clean irrigation water, reducing reliance on synthetic fertilizers and pesticides with metal contaminants, and encouraging crop rotation with phytoremediation species. Genetic screening for metal-resistant or low-accumulating wheat varieties should also be intensified, supported by national breeding programs and policy interventions. Moreover, post-harvest strategies such as proper storage and fungal contamination control can further minimize cumulative toxic exposures, especially since mycotoxins often co-occur with heavy metals and exacerbate toxicity (27,28). Future research should delve into mechanistic studies on metal transport pathways in wheat and explore the influence of soil microbial communities on metal uptake. There is also a need to evaluate the synergistic effects of heavy metals and emerging contaminants such as microplastics and persistent organic pollutants in agroecosystems. Developing region-specific safety guidelines based on cumulative risk assessment models will be crucial for ensuring food security and public health in the face of intensifying environmental stressors.

CONCLUSION

This study concludes that wheat cultivated in the Multan region contains varying concentrations of heavy metals such as cadmium, lead, iron, and zinc, reflecting environmental contamination influenced by industrial activity, fertilizer use, and soil conditions. While iron and zinc serve essential physiological roles, their excessive accumulation, alongside toxic elements like cadmium and lead, poses significant health risks, including organ damage and developmental impairments. The findings emphasize the urgent need for sustainable agricultural practices, including the use of clean irrigation water, routine soil monitoring, and the development of low-accumulating wheat varieties, to safeguard food safety and public health.



AUTHOR CONTRIBUTION

Author	Contribution		
Muhammad Haama	Substantial Contribution to study design, analysis, acquisition of Data		
Aslam*	Substantial Contribution to study design, analysis, acquisition of Data Manuscript Writing		
	Has given Final Approval of the version to be published		
	Substantial Contribution to study design, acquisition and interpretation of Data		
Esha Aslam	Critical Review and Manuscript Writing		
	Has given Final Approval of the version to be published		
Muhammad	Substantial Contribution to acquisition and interpretation of Data		
Zeeshan	Has given Final Approval of the version to be published		
Ghulam Husnain	Contributed to Data Collection and Analysis		
Ollulaili Husilaili	Has given Final Approval of the version to be published		

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