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Assessing the Bioaccumulation of Heavy Metals in Cabbage Grown under Five Soil Amendments

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Abstract: Increased heavy metal pollution worldwide necessitates urgent remediation measures. Phytoremediation stands as an eco-friendly technique that addresses this issue. This study aimed to investigate the applicability of phytoremediation in agricultural practices. Specifically, to evaluate the impact of five soil amendments (chicken manure, sewage sludge, leaf compost, cow manure, and vermicompost) on three cabbage (*Brassica oleracea* var. *capitata*) varieties (Capture, Primo vantage, and Tiara) yield, quality, and the accumulation of Cd, Cu, Mo, Ni, Pb, and Zn in cabbage heads. The bioaccumulation efficiency of cabbage was determined using an inductively coupled plasma–optical emission spectrometer (ICP-OES). Analysis revealed that soil enriched with chicken manure exhibited the highest cabbage yield. Each cabbage variety demonstrated very high bioaccumulation factor (BAF) indicating substantial heavy metal accumulation. These findings underscore the potential of utilizing crops for phytoremediation to mitigate heavy metal pollution. Additionally, the concentrations of metals below the permissible limits suggest that employing crops for phytoremediation can simultaneously ensure food productivity. This study emphasizes the necessity for further research into the use of crops for remediation strategies.

Keywords: phytoremediation; pollution; permissible limit; metal toxicity



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1. Introduction

Heavy metals are found in many places around us that can contaminate our food. They are distinct from natural contaminants and are more resistant to chemical degradation. Heavy metals have properties that make them a serious environmental threat, such as toxicity, persistence, and nonbiodegradability. This is especially true in urban areas, where contamination by heavy metals can be widespread [1]. In recent decades, heavy metal pollution in cultivated soil resulting from anthropogenic activities such as mining, smelting, manufacturing, pesticides, sewage sludge, and sewage irrigation has become a significant environmental concern for both human health and the agriculture business [2].

Heavy metals such as cadmium (Cd) and lead (Pb) can be toxic even at low concentrations. Essential heavy metals like Cu and Zn are required in small amounts in the human body, but they can also become toxic when their concentration exceeds the threshold. The essentiality and toxicity of some elements have a narrow window. It is a well-known principle in toxicology that excessive exposure to any substance can be harmful [3].

The metal uptake by plants is influenced by the bioavailability of the metal in the water phase, which is determined by its interaction with other elements and substances and retention time. Additionally, various factors such as the metal's association with soil, pH, redox potential, and soil organic matter content play a crucial role in metal existence in an ionic and plant-available form. Plants have the potential to modify the sediment's pH and oxygenation, leading to changes in soil condition and metal content [4]. In addition, the plant uptake of metals is influenced by other various factors such as soil composition, plant age, and species. Soil organic matter is crucial for soil quality and nutrient availability, and

the long-term application of NPK fertilizers and organic manure can enhance it, leading to increased levels of essential nutrients and trace metals [5–8]. On the other hand, livestock manure is a significant source of trace elements that can be taken up by plants and pose health hazards to consumers [9–12].

Organic matter in soil amendments can influence the behavior of heavy metals in the soil by releasing heavy metals associated with the organic matter, extracting, or mobilizing heavy metals from the complexes, and improving soil microbial populations. Soil microbial populations can affect heavy metal mobility and availability to the plant through the release of chelating agents, acidification, phosphate solubilization, and redox changes [13].

The type of feed consumed by poultry affects the metal content in their feces, which can contaminate the soil and the growing plants. Mineral additives in animal feed and fertilizers can also contain potentially toxic metals such as Cd and Pb, which can elevate their levels in soils and plants [14]. The accumulation of metals in the soil can also be attributed to the use of fertilizers (mineral and organic) that contain trace elements, which are essential for plant growth but can also accumulate in the soil over time [15]. These metals can be easily taken up by plants and become a part of the food chain [16].

The mineral content of vegetables is affected by the natural trace elements present in their environment, as well as the levels of minerals found in fertilizers and fertilizer doses. Heavy metals can be naturally sourced from bedrock or introduced to soils through organic and mineral fertilizers, plant protection products, industrial sewage, or road traffic pollution. Cd is the most hazardous environmental contaminant due to its poisonous and mutagenic effects on both plants and animals [17]. Furthermore, Cd is easily absorbed by plants and subsequently transmitted into human bodies via the food chain, posing a public health danger [18]. The effects of Cd on plants have been extensively researched. Cd buildup not only stops plants from growing [19] but also causes physiological and biochemical alterations [20]. Cd can disrupt photosynthetic activity, chlorophyll production, water relations, and mineral absorption; impede metabolic activities; and cause oxidative stress once it enters the plant system [21].

Vegetables grown near industrial plants, busy roads, and agricultural wastewater have elevated levels of certain elements. This can impact their availability for absorption by the human body and their interactions with other food ingredients and metals in the human body [22,23].

Some plants can extract, sequester, or detoxify heavy metals from contaminated soils, but their effectiveness depends on factors such as soil composition, metal mobility, plant factors, and crop management. Agronomic interventions such as increasing above-ground biomass, adding organic materials, intercropping, and including legumes can enhance phytoextraction through *Brassica* species by promoting growth and soil metal dissolution [24].

Plants suited for phytoremediation techniques have been the subject of much investigation. Although several *Brassica* species have been reported to be appropriate for this ecologically friendly approach that has a moderate-to-high resistance to a variety of heavy metals, it appears that the technique's full potential has yet to be realized. The cultivation of heavy metal-tolerant plant species suitable for the heavy metal remediation of polluted areas may offer a promising approach [25].

The objectives of this study were to (1) assess the effect of five soil amendments (chicken manure, sewage sludge, leaf compost, cow manure, and vermicompost) on cabbage yield and head quality and (2) assess the effect of five soil amendments on the accumulation of Cu, Cd, Pb, Ni, Mo, and Zn in cabbage heads.

2. Materials and Methods

2.1. Cabbage Field and Experimental Design

The research design used was a split block design with a total of 18 plots (3 replications × 6 treatments). Each plot had a dimension of 0.93 m × 0.93 m. Five soil amendments were used: sewage sludge (obtained from Metropolitan Sewer District, Louisville, KY, USA);

vermicompost (obtained from Wiggle Worm Soil Builder, Union Grove, WI, USA); cow manure (Lowe's, Frankfort, KY, USA); chicken manure (obtained from Alltech Chicken Facility, Lexington, KY, USA); and leaf compost (from C & R Mulch, Lexington, KY, USA) along with the control treatment (native soil). The native soil in the experimental plots was Bluegrass–Maury silty loam with 56% silt, 38% clay, and 6% sand. The soil had a pH of 6.1; an organic matter content of 2.2%; and total metal concentrations of Cu 10.17 mg/kg, Cd 0.23 mg/kg, Pb 31.2 mg/kg, Mo 0.6435 mg/kg, Zn 59.5 mg/kg, and Ni 17.15 mg/kg.

Soil amendments were applied at five percent N to eliminate variations in cabbage yield due to the N content. Amendments in each treatment (except the control) were applied to native soil to a depth of 15 cm of topsoil (Table 1). The native soils in three plots were used as a no-mulch (NM) control treatment (roto-tilled bare soil) for comparison purposes. Three cabbage (*Brassica oleracea* var. *capitata*) varieties—Capture, Primo vantage, and Tiara—were planted. Six-week-old cabbage seedlings were transplanted in the prepared field layout to a depth of 1.5 inches at 2 ft. row and plant spacing. The plants were irrigated through a uniform drip irrigation system and weeded during the growing season following The Kentucky Vegetable Grower's Guide [26]. All other cultivation practices were carried out as per the University of Kentucky Grower's Guide. The growing plants were sprayed with the insecticide esfenvalerate (Asana XL) (Valent Biosciences, Libertyville, IL, USA) three times during the growing season at one-week intervals for insect control.

Table 1. Rate of soil amendments incorporated in experimental plots.

Soil Amendments	Rate (g/m ²)
Vermicompost (Vermi.)	1120.52
Sewage sludge (SS)	224.54
Chicken manure (CM)	1022.57
Cow manure (Cow)	1937.5
Leaf compost (Leaf)	322.92

2.2. Data Collection and Soil Sampling

At harvest, three plants from each row were collected at random from each of the 18 field plots (six replicates for each soil treatment). Cabbage was harvested two times on 30 September 2022 and 3 November 2022. The harvested cabbage heads were weighed and graded according to the United States Department of Agriculture (USDA) Standards for grades of cabbage into US No. 1, US commercial, and Cull [27].

Cabbage US No. 1 consists of heads that are of reasonable solidity; not puffy or burst; and free from rot or damage caused by discoloration, disease, insects, or mechanical damage. US commercial meets all the requirements of the US No. 1 grade except for the increased tolerance for defects and that the heads are reasonably firm. Cull consists of cabbage that fails to meet the requirements of US commercial.

Native and amended soil were collected to a depth of 15 cm from field plots using a soil core sampler equipped with a plastic liner (Clements Associates, Newton, IA, USA). The sampling was performed at the time of each harvest. Soil samples were air-dried in an oven at 65 °C for 48 h, ground with mortar and pestle, and sieved through a nonmetal sieve to a size of 2 mm and kept in plastic bags [28].

2.3. Metal Analysis

2.3.1. Quantification of Metals in Harvested Plants

For the quantification of metals in harvested plants, cabbage heads of appropriate size were randomly collected from each plot. Soils were carefully removed from the cabbage heads and washed with deionized water to remove any attached soil particles. Cabbage heads were then dried in an oven at 65 °C for 48 h. Using ceramic mortar and pestle, the dried samples were manually ground to pass a 2 mm nonmetal sieve. Samples were

re-dried using an oven to obtain constant weight. Then, 5 mL of concentrated nitric acid (HNO₃) trace metal grade was added to 0.5 g of each dry sample powder, and the mixture was digested on a Digi block digestion system at 95 °C for 1.5 h. A final digestion procedure of 4 mL of 30% peroxide was carried out for 30 min. The mixture was then diluted with deionized water to 50 mL [29,30]. Metal concentration was determined using an inductively coupled plasma–optical emission spectrometer (ICP-OES) in standard mode following the SW-846 EPA 6010 B method [31]. The apparatus was calibrated using a multielement standard (TruQ 500 mL, PerkinElmer, Waltham, MA, USA). Various working dilutions (at concentrations of 1, 10, 20, and 50 ppm) of the analytical standard were prepared to create calibration curves. Additionally, a standard solution with a concentration of 10 ppm served as a check standard to ensure accuracy. The acceptable outcome was assessed to be within a 10% margin. Each sample was measured in three replications, and the result was calculated as the arithmetic mean, with a mean difference of less than 5%. Instrument detection limits (IDLs) were 0.0027 mg/kg for Cd, 0.0054 mg/kg for Cu, 0.014 mg/kg for Mo, 0.048 mg/kg for Ni, 0.042 mg/kg for Pb, and 0.0018 mg/kg for Zn.

2.3.2. Evaluation of Soluble Concentrations of Metals in Soil

Soluble concentrations of metals in soil were extracted using calcium chloride (CaCl₂) to quantify soluble and extractable metals in soil. Soil samples (10 g) were suspended in 25 mL of 0.01 CaCl₂ and heated at 90 °C for 30 min. The resulting supernatants were filtered while hot through Whatman filter paper #42, and 2 drops of 1 M HNO₃ trace metal grade were added to prevent metal precipitation and inhibit microbial growth in sample extracts [32,33]. Finally, the samples were analyzed using ICP spectrometry.

2.4. Bioaccumulation Factor (BAF)

The bioaccumulation factor for heavy metals was calculated as given by Khan et al. [34] and Mirecki et al. [35].

$$BAF = \frac{\text{Soluble concentration of heavy metals in crops}}{\text{Soluble concentration of heavy metals in soil}}$$

2.5. Statistical Analysis

The obtained data were statistically analyzed using one-way ANOVA. The assumption was that the soil treatments influence the yield (response variable) of the three varieties of cabbage and the mobility of heavy metals from soil to cabbage. Initially, the response variables were plotted to visualize the distribution of the data. The normality of the data was tested using the Shapiro–Wilk test in R [36]. The mean group comparison among the treatments was calculated using the ‘multcomp::cld’ function in the R package. The data were analyzed using the R programming language for computing and analysis [37].

3. Results and Discussions

3.1. Yield and Quality

The total cabbage head yield obtained from the soil amended with chicken manure was significantly higher ($p \leq 0.05$) than the yield obtained from the leaf compost but not significantly different from the other treatments (Figure 1). This could be due to the composition of chicken manure, which is a mixture of feces, waste feed, feathers, and bedding materials that contains NPK at approximately 8.5% of the weight of the manure [38]. Antonious et al. [28] also found that soil amended with chicken manure had significantly higher cabbage head yield than the control treatment.

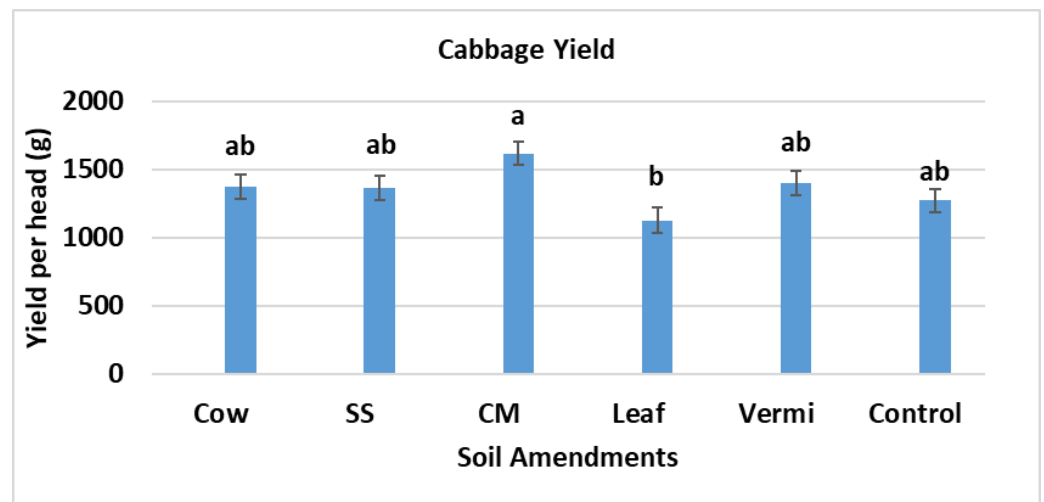


Figure 1. Total yield of cabbage grown under six soil management practices (Cow = cow manure, SS = sewage sludge, CM = chicken manure, Leaf = leaf compost, Vermi = vermicompost, and Control = native soil). Bars accompanied by different letter(s) are significantly different ($p \leq 0.05$) from each other. Samples were analyzed using ‘multicomp::cld’ function from R 2023.

When comparing the varieties, the cabbage head yield obtained from the Primo vantage variety was significantly higher ($p \leq 0.05$) than those obtained from the Tiara and Capture varieties (Figure 2). Similarly, the US 1 cabbage grade had a significantly higher ($p \leq 0.05$) yield than the US commercial and Cull varieties (Figure 3). The good quality and quantity of the harvest can be due to the addition of soil amendments that increase the availability of nutrients to plants and enhance the organic matter, water-holding capacity, total pore space, and soil aggregate stability [39].

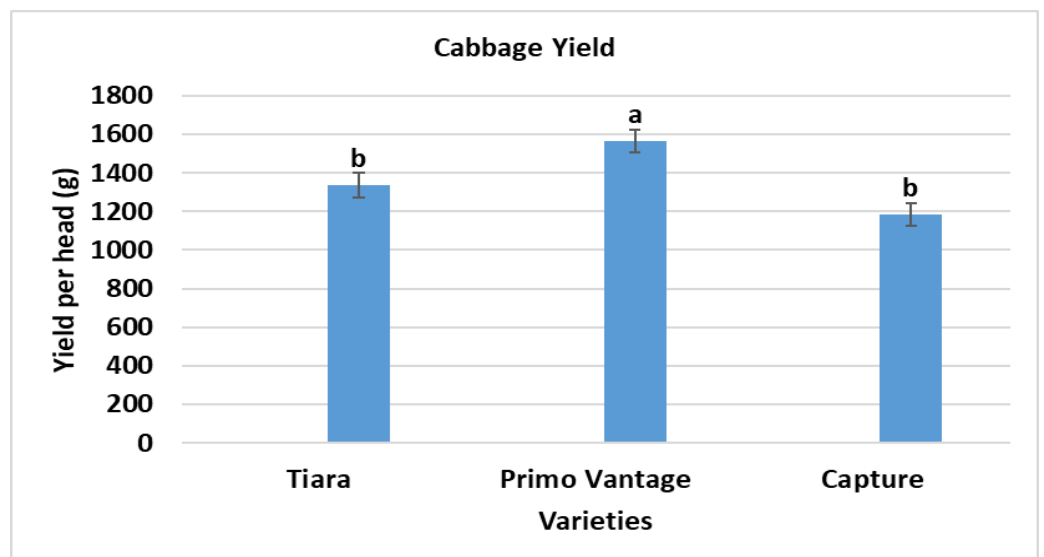


Figure 2. Total yield of cabbage grown under six soil management practices. Statistical comparisons were carried out among six soil management practices for each variety. Bars accompanied by different letter(s) are significantly different ($p \leq 0.05$) from each other. Samples were analyzed using ‘multicomp::cld’ function from R 2023 (R Core Team, 2023).

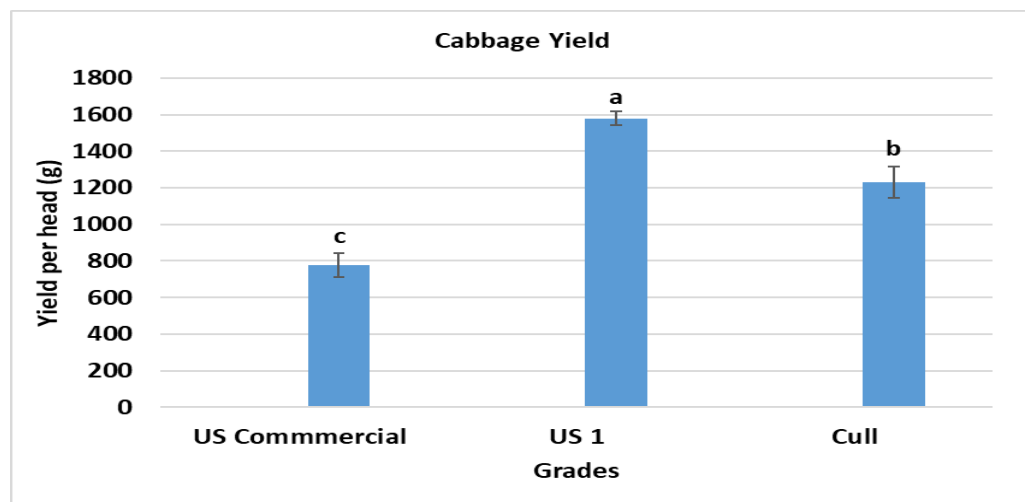


Figure 3. Variability in cabbage grades of plants grown under six soil management practices. Statistical comparisons were carried out among three cabbage grades. Bars accompanied by different letter(s) are significantly different ($p \leq 0.05$) from each other. Samples were analyzed using 'multcomp::cld' function from R 2023.

The utilization of soil amendments in commercial agriculture offers an economical means to enhance both crop yield and fruit quality, especially benefiting limited-resource farmers. However, there is a crucial need for field research that aims to determine the ideal application rate of animal manure fertilizer within agricultural production systems, focusing on its impact on both crop yield and the quality of marketable fruit [40].

3.2. Total and Soluble Heavy Metal Concentrations in Soil

The total metal concentration (extracted with HNO_3) was higher than the soluble metal concentration (extracted with CaCl_2) for all the quantified metals. The average concentration of total metal in soil decreased in the sequence of $\text{Zn} > \text{Pb} > \text{Ni} > \text{Cu} > \text{Mo} > \text{Cd}$, whereas the soluble metal concentration in soil decreased in the sequence of $\text{Zn} > \text{Cu} > \text{Pb} > \text{Ni} > \text{Mo} > \text{Cd}$.

The total metal concentration for all the metals in the soil (Table 2) was below the permissible limit by the FAO and WHO [41] (Table 3).

3.3. BAF Values of Cabbage var. Primo Vantage

The soil amended with cow manure showed the highest bioaccumulation factor (BAF) value for Cu, which was significantly higher than the control treatment. The soil amended with leaf compost showed the highest Pb BAF value, which was significantly higher than the control treatment. Similarly, the soil amended with the vermicompost showed the highest BAF value for Cd, which was significantly different from the leaf compost. For Mo, the cow manure and sewage sludge showed the highest BAF values, which were significantly higher than the control treatment. The sewage sludge showed the highest BAF value for Ni, which was significantly higher than the chicken manure. The sewage sludge and leaf compost showed the highest BAF values for Zn, which were significantly higher than the control (Figure 4).

Table 2. Soluble metal concentration in experimented soil.

Amendments	Metals	Harvest 1		Harvest 2	
		Total Metal Content (mg/kg)	Soluble Metal Content (mg/kg)	Total Metal Content (mg/kg)	Soluble Metal Content (mg/kg)
SS	Cd	0.237 ± 0.02	0.015 ± 0.00	0.22 ± 0.26	0.015 ± 0.00
	Cu	9.63 ± 0.09	0.05 ± 0.009	9.52 ± 0.16	0.05 ± 0.009
	Mo	0.657 ± 0.08	0.015 ± 0.00	0.640 ± 0.07	0.015 ± 0.00
	Ni	17.2 ± 0.93	0.015 ± 0.001	17.1 ± 0.23	0.015 ± 0.001
	Pb	30.4 ± 1.08	0.036 ± 0.01	29.1 ± 0.3	0.036 ± 0.01
	Zn	57.4 ± 3.17	0.156 ± 0.05	57.2 ± 1.67	0.156 ± 0.05
Cow manure	Cd	0.253 ± 0.05	0.015 ± 0.00	0.243 ± 0.01	0.015 ± 0.00
	Cu	9.55 ± 0.45	0.03 ± 0.009	9.45 ± 0.56	0.03 ± 0.009
	Mo	0.673 ± 0.07	0.015 ± 0.00	0.660 ± 0.05	0.015 ± 0.00
	Ni	16.4 ± 0.73	0.016 ± 0.001	16.1 ± 0.53	0.016 ± 0.001
	Pb	28.9 ± 1.30	0.02 ± 0.01	28.7 ± 1.53	0.022 ± 0.01
	Zn	56.7 ± 5.21	0.125 ± 0.05	53.5 ± 3.38	0.125 ± 0.05
Vermicompost	Cd	0.317 ± 0.01	0.015 ± 0.00	0.24 ± 0.02	0.015 ± 0.00
	Cu	9.86 ± 0.77	0.04 ± 0.009	9.7 ± 0.16	0.04 ± 0.009
	Mo	0.847 ± 0.06	0.015 ± 0.00	0.740 ± 0.03	0.015 ± 0.00
	Ni	17.1 ± 0.88	0.015 ± 0.001	16.8 ± 0.50	0.015 ± 0.001
	Pb	30.9 ± 1.29	0.03 ± 0.01	29.7 ± 0.65	0.03 ± 0.01
	Zn	56.4 ± 3.9	0.055 ± 0.05	55.6 ± 1.38	0.055 ± 0.05
Leaf compost	Cd	0.327 ± 0.01	0.015 ± 0.00	0.263 ± 0.02	0.015 ± 0.00
	Cu	9.66 ± 0.80	0.02 ± 0.009	9.46 ± 0.76	0.024 ± 0.009
	Mo	0.830 ± 0.05	0.015 ± 0.00	0.747 ± 0.18	0.015 ± 0.00
	Ni	17.4 ± 1.43	0.02 ± 0.001	17.2 ± 1.37	0.02 ± 0.001
	Pb	29.9 ± 1.85	0.03 ± 0.01	29.0 ± 2.05	0.03 ± 0.01
	Zn	55.0 ± 3.95	0.06 ± 0.05	54.2 ± 3.78	0.06 ± 0.05
Chicken manure	Cd	0.240 ± 0.02	0.015 ± 0.00	0.21 ± 0.02	0.015 ± 0.00
	Cu	10.23 ± 0.69	0.03 ± 0.009	10.10 ± 0.79	0.04 ± 0.009
	Mo	0.693 ± 0.13	0.015 ± 0.00	0.594 ± 0.11	0.015 ± 0.00
	Ni	17.3 ± 1.30	0.016 ± 0.001	17.0 ± 0.90	0.016 ± 0.001
	Pb	31.7 ± 1.97	0.03 ± 0.01	30.7 ± 0.72	0.036 ± 0.01
	Zn	60.3 ± 4.56	0.11 ± 0.05	58.7 ± 4.84	0.11 ± 0.05
Control	Cd	0.230 ± 0.00	0.015 ± 0.00	0.20 ± 0.02	0.015 ± 0.00
	Cu	10.18 ± 0.56	0.03 ± 0.009	10.06 ± 0.67	0.032 ± 0.009
	Mo	0.76 ± 0.06	0.015 ± 0.00	0.673 ± 0.11	0.015 ± 0.00
	Ni	17.3 ± 1.15	0.017 ± 0.001	17.2 ± 0.52	0.017 ± 0.001
	Pb	30.0 ± 1.56	0.03 ± 0.01	29.0 ± 0.76	0.037 ± 0.01
	Zn	55.4 ± 3.34	0.108 ± 0.05	54.6 ± 3.58	0.1 ± 0.05

Each value represents an average of three replicates of soil amendments ± std. error.

Table 3. Comparison of metal concentration from the current study with the permissible limit of heavy metals in unpolluted soil and vegetables.

Heavy Metals	Permissible Limit in Unpolluted Soil (mg/kg)	Permissible Limit in Vegetables (mg/kg)	Total Metal Concentration in Soil from the Current Study (mg/kg)	Soluble Metal Concentration in Cabbage from Current Study (mg/kg)
Cd	3	0.1	0.250	0.093
Cu	100	73	9.5	3.008
Mo	NA	NA	0.650	0.015
Ni	50	67	16.5	0.392
Pb	100	0.3	30	0.424
Zn	300	100	56	26.308

NA = not available (source: FAO and WHO, 2014).

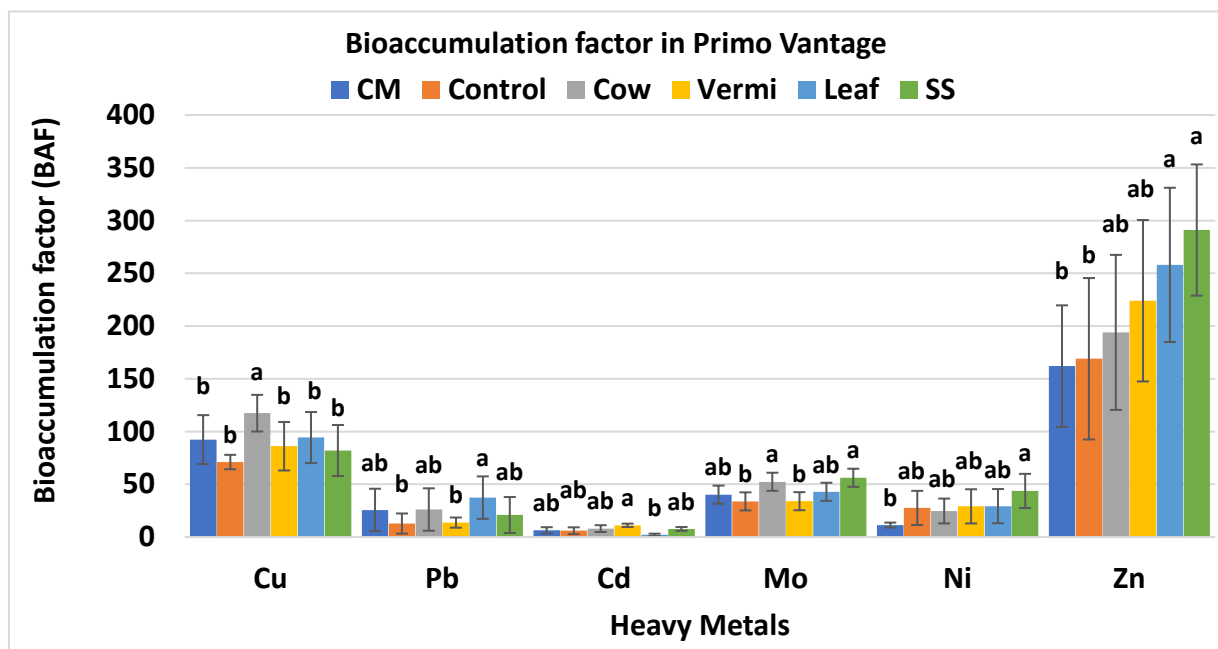


Figure 4. Bioaccumulation factor of soluble heavy metals in cabbage var. Primo vantage grown under six soil management practices (CM = chicken manure, Cow = cow manure, Vermi = vermicompost, Leaf = leaf compost, SS = sewage sludge, and Control = native soil) extracted using CaCl_2 . Bars accompanied by different letter(s) are significantly different ($p \leq 0.05$) from each other. Samples were analyzed using ‘multicomp::cld’ function from R 2023.

Studies have demonstrated that certain vegetables, classified as hyper-accumulators, can accumulate high levels of heavy metals from contaminated soils. These vegetables tend to absorb and accumulate higher concentrations of heavy metals when cultivated in metal-contaminated soil compared to uncontaminated soil, primarily through their root systems. Specifically, certain *Brassica* species, such as cabbage, are known to hyper-accumulate heavy metals in their edible tissues [42].

3.4. BAF Values of Cabbage var. Tiara

The soil amended with cow manure showed the highest BAF value for Cu, which was significantly higher than the vermicompost and sewage sludge. For Pb, the soil amended with the vermicompost showed the highest BAF value, which was significantly different from the soil amended with cow manure and leaf manure. Except for the sewage sludge, all soil amendments showed significantly higher Cd BAF values than the control treatment. There were no significant differences in BAF values for Mo among the soil treatments. Similarly, the soil amended with the chicken manure showed the highest BAF value for Ni, which was significantly higher than the control treatment, whereas the soil amended with the cow manure had a significantly higher BAF than the soil amended with the leaf compost and sewage sludge for Zn (Figure 5).

The root of a plant serves two primary functions: anchoring the plant in the soil and absorbing water and dissolved minerals. In cabbage plants, the roots play a crucial role in supplying essential nutrients and storing minerals. Additionally, cabbage roots can accumulate heavy metals, providing protection for the above-ground plant parts. The adsorption and transport of heavy metals within cabbage depends on the metal type, its cabbage-related functions, and its capacity to form complexes with sap components [43].

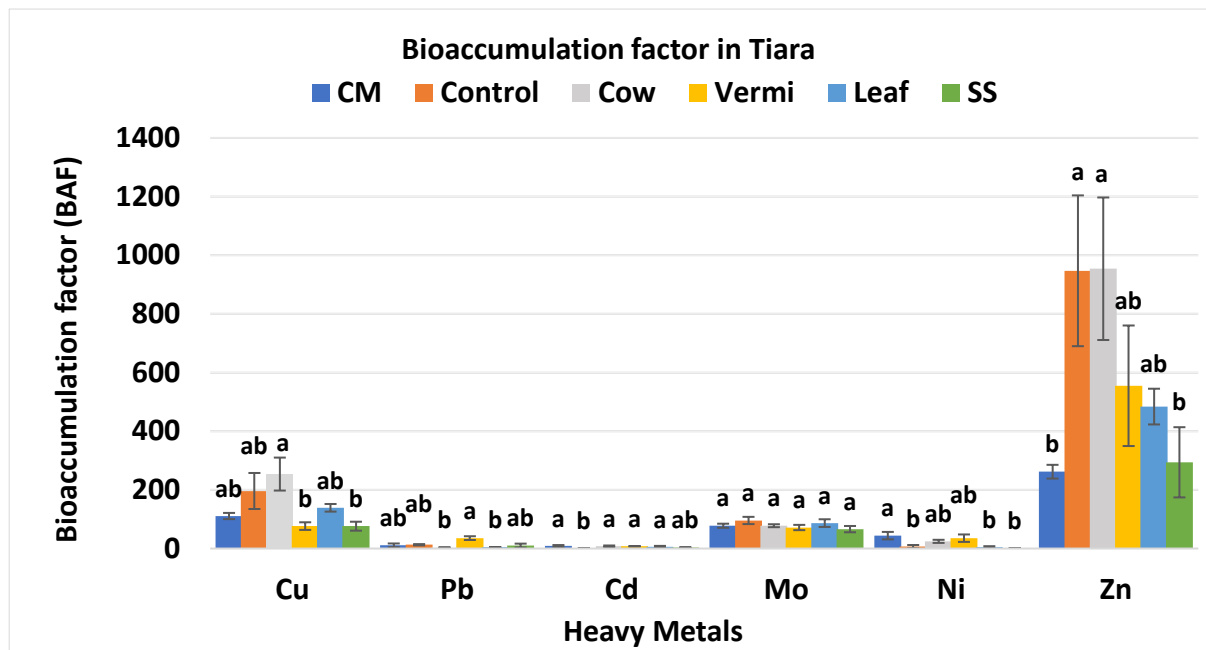


Figure 5. Bioaccumulation factor of soluble heavy metals in cabbage var. Tiara grown under six soil management practices (CM = chicken manure, Cow = cow manure, Vermi = vermicompost, Leaf = leaf compost, SS = sewage sludge, and Control = native soil) extracted using CaCl_2 . Bars accompanied by different letter(s) are significantly different ($p \leq 0.05$) from each other. Samples were analyzed using ‘multcomp::cld’ function from R 2023.

3.5. BAF Values of Cabbage var. Capture

The soil amended with leaf compost showed the highest BAF for Cu, which was significantly higher than the control treatment. For Pb, Cd, and Mo, there were no significant differences in BAF values among the soil treatments. The soil amended with cow manure showed the highest BAF value for Ni, which was significantly higher than the chicken manure, vermicompost, and sewage sludge. The soil amended with leaf compost had a significantly higher BAF than the control treatment (Figure 6).

Plants obtain heavy metals from the soil through a range of mechanisms, such as ionic exchange, adsorption, redox reactions, dissolution, and precipitation [44]. The modest accumulation of cadmium (Cd) in cabbage, as depicted in Figures 4–6, can be linked to the presence of zinc (Zn) in these plants. Earlier research has proposed that the presence of Zn can impede the adsorption of Cd, leading to a decrease in Cd accumulation within plants [45].

3.6. Overall BAF Values of Three Varieties of Cabbage

The soil amended with the chicken manure, cow manure, leaf compost, and sewage sludge treatments showed the highest BAF values for Pb, which were significantly higher than the control treatment. For Cu, Cd, Mo, and Zn BAF values, there were no significant differences ($p \geq 0.5$) among all the soil treatments. The soil amended with the sewage sludge showed the highest BAF for Ni, which was significantly higher than the chicken manure (Figure 7).

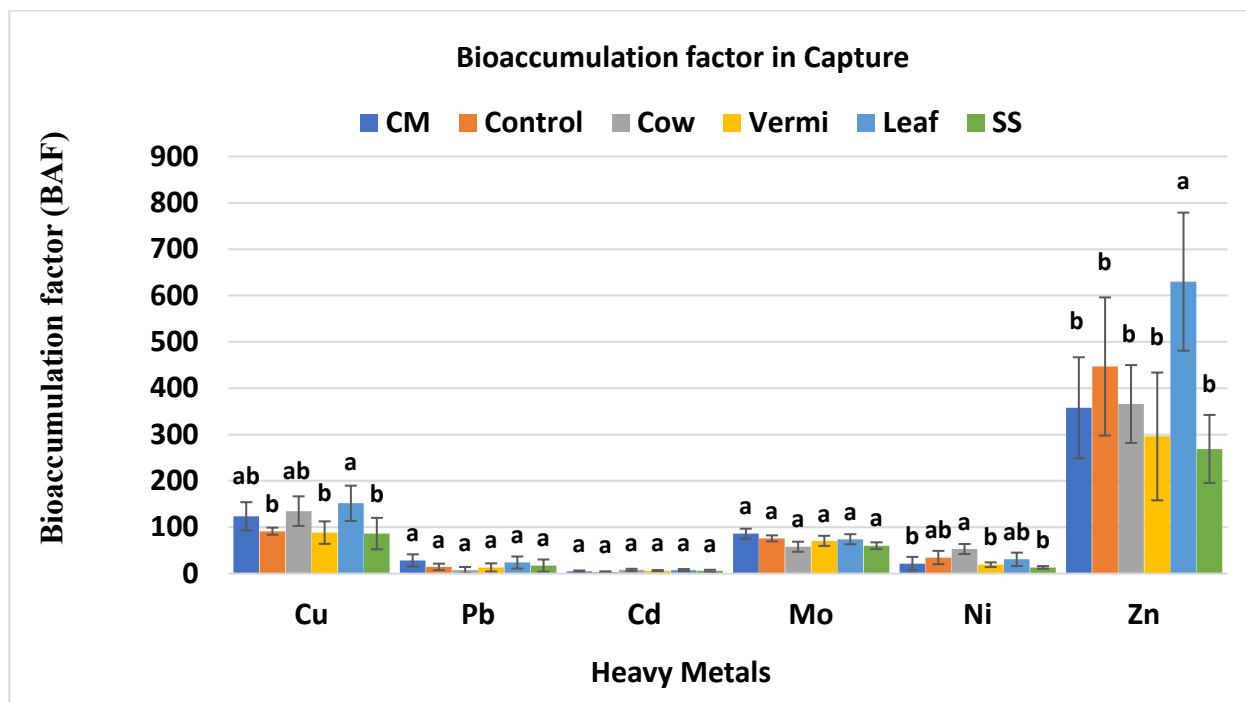


Figure 6. Bioaccumulation factor of soluble heavy metals in cabbage var. Capture grown under six soil management practices (CM = chicken manure, Cow = cow manure, Vermi = vermicompost, Leaf = leaf compost, SS = sewage sludge, and Control = native soil) extracted using CaCl_2 . Bars accompanied by different letter(s) are significantly different ($p \leq 0.05$) from each other. Samples were analyzed using ‘multcomp::cld’ function from R 2023.

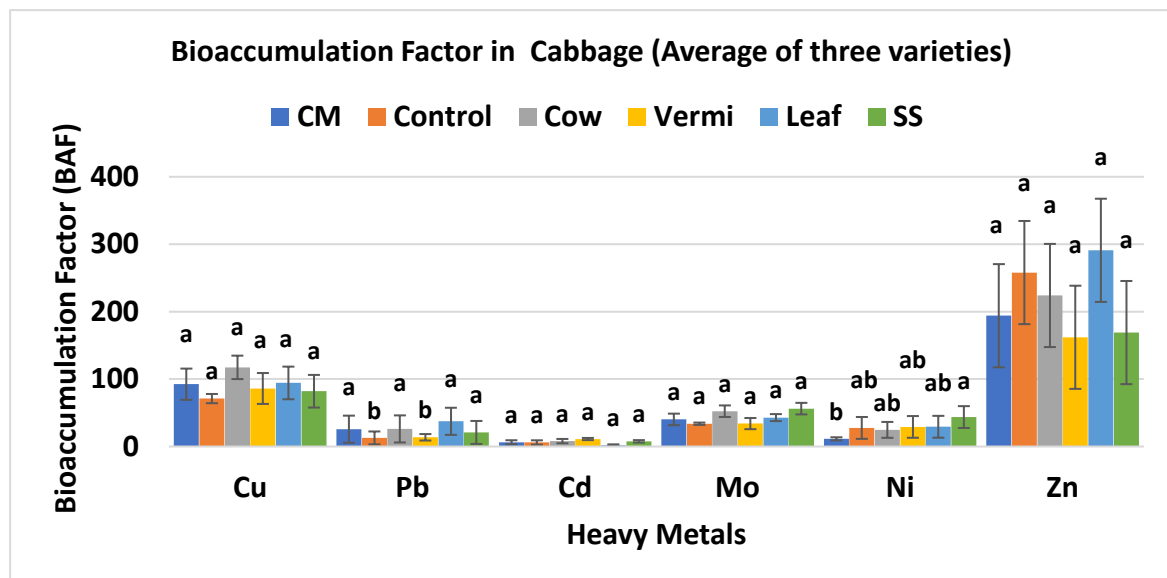


Figure 7. Overall bioaccumulation factor of soluble heavy metals of three varieties of cabbage grown under six soil management practices (CM = chicken manure, Cow = cow manure, Vermi = vermicompost, Leaf = leaf compost, SS = sewage sludge, and Control = native soil) extracted using CaCl_2 . Bars accompanied by different letter(s) are significantly different ($p \leq 0.05$) from each other. Samples were analyzed using ‘multcomp::cld’ function from R 2023.

The BAF values of heavy metals for cabbage revealed an increasing order of $\text{Cd} < \text{Pb} < \text{Ni} < \text{Mo} < \text{Cu} < \text{Zn}$. BAF values for all the heavy metals tested in cabbage were very high ($\text{BAF} \gg 1$), which means that cabbage can be used to accumulate these heavy metals

from the soil for bioremediation purposes. The heavy metal concentration in cabbage tissues was below the permissible limit by the FAO and WHO (2014) for all heavy metals except for Pb (Table 2). The mean concentration of Pb in cabbage was 0.42 mg/kg, which was above the permissible limit of 0.30 mg/kg. This means that cabbage can show Pb toxicity when consumed by humans. A study from Pajević et al. [46] in Serbia reported that the accumulation of Cd, Pb, and Ni were 0.89 mg/kg, 3.56 mg/kg, and 2.2 mg/kg, respectively, in the edible parts of cabbage. Similarly, Ametepey et al. [47] also reported that the accumulation of Cd and Pb were 0.01–0.007 and 0.05 mg/kg, respectively, in the edible parts of cabbage. Arora et al. in [48] found that Cu (10–73.8 mg/kg) and Zn (4.8–22.5 mg/kg) were accumulated in the edible parts of cabbage. In addition, the results showed that the accumulation of Cd was much lower than that of the other heavy metals. According to Riaz et al. [49], the solubility of Cd in soil solution as well as its uptake by plants are strongly influenced by the amount of organic matter in the soil. This means that soils with higher organic matter content may limit the uptake of Cd by plants.

Similarly, the accumulation of Pb was also low. A study reported that Pb is poorly mobile in contaminated soil and is rarely present in the form of Pb^{2+} but forms complex ions $PbOH^+$ and $Pb(OH)_4^{2-}$ that regulate sorption and desorption processes. It is strongly bound by most soil components like iron (Fe) and manganese (Mn) [50]. The accumulation of Zn was the highest among the metals. This might be due to the formation of complex ions of Zn in the soil, which dissolves easily under weathering processes, making it highly mobile and easy to accumulate in the surface horizons of mineral and organic soils. Also, organic matter forms stable bonds with Zn, increasing its mobility, and adding municipal waste to soil increases its mobile forms [51].

A distinguishing aspect of this study lies in its pioneering finding, specifically in highlighting that crops employed for phytoremediation purposes can concurrently serve as edible produce. This duality stems from the observation that the quantified concentrations of heavy metals in cabbage remain within permissible limits, except for Cd and Pb. These outcomes hold significant implications for both environmental sustainability and agricultural productivity. They underscore the potential to not only effectively mitigate heavy metal pollution in soils but also ensure the safety of food products. This dual-purpose approach, which addresses both environmental concerns and food safety, adds a unique dimension to the field of research.

The present study demonstrates the efficiency of cabbage in accumulating elevated levels of Zn and Cu while exhibiting lower concentrations of Cd, Mo, Ni, and Pb from the soil, thereby showcasing its potential as a phytoremediation candidate. Consequently, further investigations are needed to explore the full extent of the phytoremediation capabilities of cabbage and crops like cabbage. These studies should also encompass the bioabsorption potential of cabbage for other toxic heavy metals. Additionally, research should delve into various soil-related aspects, including the correlation between soil pH and the mechanisms governing metal uptake by plants, to enhance our comprehension of heavy metal bioaccumulation.

4. Conclusions

Cabbage is consumed in the US and all around the world. Among the cabbage varieties, the Primo vantage variety had the highest bioaccumulation of heavy metals, followed by the Capture variety. The metal concentration extracted using nitric acid was higher than that extracted using the calcium chloride solution. This leads us to the conclusion that the high level of a metal concentration in soils does not necessarily reflect the metal mobility to plants. Plants accumulate heavy metals from soils at different levels depending on plant species and genotype within the same species, soil pH, and organic matter content.

The BAF value of heavy metals in cabbage was very high ($BAF \gg 1$), which indicates that the cabbage is a good accumulator of Cu, Cd, Ni, Mo, and Zn. This shows that cabbage can be used for the phytoremediation of heavy metals. Cabbage accumulated Cd, Cu, Ni, and Zn concentrations below the permissible limit, whereas the Pb concentration was

above the permissible limit. This shows there is a risk of Pb toxicity when cabbage is grown and consumed under these field conditions. One should avoid Pb toxicity that can occur through the ingestion of food crops grown in soils with high Pb concentrations.

The elevated level of Pb in cabbage grown in soils mixed with the five soil amendments showed that growers should avoid using vermicompost and cow manure when growing cabbage on sites having high levels of Pb. We recommend using the Primo vantage variety of cabbage to avoid heavy metal toxicity. A well-planned approach is required to reduce the risk associated with the use of food crops polluted with heavy metals. Bioremediation can be an environmentally friendly and cost-effective method to reduce heavy metal toxicity in polluted soils.

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References

1. Gu, Y.G.; Lin, Q.; Gao, Y.P. Metals in exposed-lawn soils from 18 urban parks and its human health implications in southern China's largest city, Guangzhou. *J. Clean. Prod.* **2016**, *115*, 122–129. [\[CrossRef\]](#)
2. Antonkiewicz, J.; Pelka, R.; Bik-Malodzinska, M.; Zukowska, G.; Glen-Karolczyk, K. The effect of cellulose production waste and municipal sewage sludge on biomass and heavy metal uptake by a plant mixture. *Environ. Sci. Pollut. Res. Int.* **2018**, *25*, 31101–31112. [\[CrossRef\]](#) [\[PubMed\]](#)
3. Ali, H.; Khan, E.; Ilahi, I. Environmental chemistry and ecotoxicology of hazardous heavy metals: Environmental persistence, toxicity, and bioaccumulation. *J. Chem.* **2019**, *2019*, 6730305. [\[CrossRef\]](#)
4. Kumar, M.; Gupta, N.; Ratn, A.; Awasthi, Y.; Prasad, R.; Trivedi, A.; Trivedi, S.P. Biomonitoring of heavy metals in river ganga water, sediments, plant, and fishes of different trophic levels. *Biol. Trace Elem. Res.* **2020**, *193*, 536–547. [\[CrossRef\]](#)
5. Khan, Z.I.; Safdar, H.; Ahmad, K.; Wajid, K.; Bashir, H.; Ugulu, I.; Dogan, Y. Health risk assessment through determining bioaccumulation of iron in forages grown in soil irrigated with city effluent. *Environ. Sci. Pollut. Res.* **2019**, *26*, 14277–14286. [\[CrossRef\]](#) [\[PubMed\]](#)
6. Nadeem, M.; Qureshi, T.M.; Ugulu, I.; Riaz, M.N.; An, Q.U.; Khan, Z.I.; Ahmad, K.; Ashfaq, A.; Bashir, H.; Dogan, Y. Mineral, vitamin and phenolic contents and sugar profiles of some prominent date palm (*Phoenix dactylifera*) varieties of Pakistan. *Pak. J. Bot.* **2019**, *51*, 171–178. [\[CrossRef\]](#) [\[PubMed\]](#)
7. Yorek, N.; Ugulu, I.; Aydin, H. Using self-organizing neural network map combined with ward's clustering algorithm for visualization of students' cognitive structural models about aliveness concept. *Comput. Intell. Neurosci.* **2016**, *2016*, 2476256. [\[CrossRef\]](#) [\[PubMed\]](#)
8. Rezig, F.A.M.; Mubarak, A.R.; Ehadi, E.A. Impact of organic residues and mineral fertilizer application on soil–crop system: II soil attributes. *Arch. Agron. Soil Sci.* **2013**, *59*, 1245–1261. [\[CrossRef\]](#)
9. Dogan, Y.; Unver, M.C.; Ugulu, I.; Calis, M.; Durkan, N. Heavy metal accumulation in the bark and leaves of *Juglans regia* planted in Artvin City, Turkey. *Biotechnol. Biotechnol. Equip.* **2014**, *28*, 643–649. [\[CrossRef\]](#)
10. Khan, Z.I.; Ahmad, K.; Safdar, H.; Ugulu, I.; Wajid, K.; Bashir, H.; Dogan, Y. Manganese bioaccumulation and translocation of in forages grown in soil irrigated with city effluent: An evaluation on health risk. *Res. J. Pharm. Biol. Chem. Sci.* **2018**, *9*, 759–770.
11. Khan, Z.I.; Ugulu, I.; Umar, S.; Ahmad, K.; Mehmood, N.; Ashfaq, A.; Bashir, H.; Sohail, M. Potential toxic metal accumulation in soil, forage and blood plasma of buffaloes sampled from Jhang, Pakistan. *Bull. Environ. Contam. Toxicol.* **2018**, *101*, 235–242. [\[CrossRef\]](#) [\[PubMed\]](#)

12. Khan, Z.I.; Ugulu, I.; Ahmad, K.; Yasmeen, S.; Noorka, I.R.; Mehmood, N.; Sher, M. Assessment of trace metal and metalloid accumulation and human health risk from vegetables consumption through spinach and coriander specimens irrigated with wastewater. *Bull. Environ. Contam. Toxicol.* **2018**, *101*, 787–795. [\[CrossRef\]](#) [\[PubMed\]](#)
13. Premi, O.P.; Kandpal, B.K.; Rathore, S.S.; Shekhawat, K.; Chauhan, J.S. Green manuring, mustard residue recycling and fertilizer application affects productivity and sustainability of Indian mustard (*Brassica juncea* L.) in Indian semi-arid tropics. *Ind. Crop. Prod.* **2013**, *41*, 423–429. [\[CrossRef\]](#)
14. Sakizadeh, M.; Faraji, F.; Pouraghniyayi, M.J. Quality of groundwater in an area with intensive agricultural activity. *Expo. Health* **2016**, *8*, 93–105. [\[CrossRef\]](#)
15. Atafar, Z.; Mesdaghinia, A.; Nouri, J.; Homaei, M.; Yunesian, M.; Ahmadimoghaddam, M.; Mahvi, A.H. Effect of fertilizer application on soil heavy metal concentration. *Environ. Monit. Assess.* **2010**, *160*, 83–89. [\[CrossRef\]](#)
16. Ugulu, I.; Unver, M.C.; Dogan, Y. Potentially toxic metal accumulation and human health risk from consuming wild *Urtica urens* sold on the open markets of Izmir. *Euro-Mediterr. J. Environ. Integr.* **2019**, *4*, 36. [\[CrossRef\]](#)
17. Huang, Y.; He, C.; Shen, C.; Guo, J.; Yang, Z. Toxicity of cadmium and its health risks from leafy vegetable consumption. *Food Funct.* **2017**, *8*, 1373–1401. [\[CrossRef\]](#)
18. Zhou, Q.; Yang, Y.; Shen, C.; He, C.; Yuan, J.; Yang, Z. Comparative analysis between low and high cadmium accumulating cultivars of *Brassica parachinensis* to identify difference of cadmium-induced microRNA and their targets. *Plant Soil* **2017**, *420*, 223–237. [\[CrossRef\]](#)
19. Rizwan, M.; Ali, S.; Abbas, T.; Zia-ur-Rehman, M.; Hannan, F.; Kellerc, C.; Al-Wabel, M.I.; Ok, Y.S. Cadmium minimization in wheat: A critical review. *Ecotoxicol. Environ. Saf.* **2016**, *130*, 43–53. [\[CrossRef\]](#)
20. Ahmad, A.; Hadi, F.; Ali, N. Effective phytoextraction of cadmium (Cd) with increasing concentration of total phenolics and free proline in *Cannabis sativa* (L) plant under various treatments of fertilizers, plant growth regulators and sodium salt. *Int. J. Phytoremediation* **2015**, *17*, 56–65. [\[CrossRef\]](#)
21. Baczek-Kwinta, R.; Juzo, K.; Borek, M.; Antonkiewicz, J. Photosynthetic response of cabbage in cadmium-spiked soil. *Photosynthetica* **2019**, *57*, 731–739. [\[CrossRef\]](#)
22. Śmiechowska, M.; Florek, A. Content of heavy metals in selected vegetables from conventional, organic and allotment cultivation. *J. Res. Appl. Agric. Eng.* **2011**, *56*, 152–156.
23. Zwolak, A.; Sarzyńska, M.; Szpyrka, E.; Stawarczyk, K. Sources of soil pollution by heavy metals and their accumulation in vegetables: A review. *Water Air Soil Pollut.* **2019**, *230*, 164. [\[CrossRef\]](#)
24. Rathore, S.S.; Shekhawat, K.; Dass, A.; Kandpal, B.K.; Singh, V.K. Phytoremediation mechanism in Indian mustard (*Brassica juncea*) and its enhancement through agronomic interventions. *Proc. Natl. Acad. Sci. India Sect. B Biol. Sci.* **2019**, *89*, 419–427. [\[CrossRef\]](#)
25. Witters, N.; Mendelsohn, R.O.; van Slycken, S.; Weyens, N.; Schreurs, E.; Meers, E.; Tack, F.; Carleer, R.; Vangronsveld, J. Phytoremediation, a sustainable remediation technology? Conclusions from a case study. I: Energy production and carbon dioxide abatement. *Biomass Bioenergy* **2012**, *39*, 454–469. [\[CrossRef\]](#)
26. Pfeufer, E.; Bessin, R.; Wright, S.; Strang, J. *Vegetable Production Guide for Commercial Growers*; Cooperative Extension Service, University of Kentucky College of Agriculture, Food and Environment: Lexington, KY, USA, 2017; pp. 44–48.
27. USDA. *United States Standards for Grades of Cabbage*; United States Department of Agriculture, Agricultural Marketing Service: Washington, DC, USA, 2016. Available online: <https://www.ams.usda.gov/grades-standards/cabbage-grades-and-standards> (accessed on 2 March 2022).
28. Antonious, G.F.; Kochhar, T.S.; Coolong, T. Yield, quality, and concentration of seven heavy metals in cabbage and broccoli grown in sewage sludge and chicken manure amended soil. *J. Environ. Sci. Health Part A* **2012**, *47*, 1955–1965. [\[CrossRef\]](#)
29. Matejovic, I.; Durackova, A. Comparison of microwave digestion, wet and dry mineralization, and solubilization of plant samples for determination of calcium, magnesium, potassium, phosphorus, sodium, iron, zinc, copper, and manganese. *Commun. Soil Sci. Plant Anal.* **1994**, *25*, 1277–1288. [\[CrossRef\]](#)
30. Lee, J.; Park, Y.S.; Lee, D.Y. Fast and green microwave-assisted digestion with diluted nitric acid and hydrogen peroxide and subsequent determination of elemental composition in brown and white rice by ICP-MS and ICP-OES. *LWT* **2023**, *173*, 11435. [\[CrossRef\]](#)
31. Environmental Protection Agency. Method 6010b Inductively Coupled Plasma Atomic Emission Spectrometry. Revision 2. 1996. Available online: <https://www.epa.gov/sites/default/files/documents/6010b.pdf> (accessed on 1 March 2023).
32. McBride, M.B.; Richards, B.K.; Steenhuis, T. Bioavailability and crop uptake of trace elements in soil columns amended with sewage sludge products. *Plant Soil* **2004**, *262*, 71–84. [\[CrossRef\]](#)
33. Antonious, G.F.; Turley, E.T.; Kochhar, T.S. Testing bioaccumulation of Cd, Pb, and Ni in plants grown in soil amended with municipal sewage sludge at three Kentucky locations. *JSM Environ. Sci. Ecol.* **2017**, *5*, 1039.
34. Khan, S.; Rehman, S.; Khan, A.Z.; Khan, M.A.; Shah, M.T. Soil and vegetables enrichment with heavy metals from geological sources in Gilgit, northern Pakistan. *Ecotoxicol. Environ. Saf.* **2010**, *73*, 1820–1827. [\[CrossRef\]](#)
35. Mirecki, N.; Agic, R.; Sunic, L.; Milenkovic, L.; Ilic, Z.S. Transfer factor as indicator of heavy metals content in plants. *Fresenius Environ. Bull.* **2015**, *24*, 4212–4219.
36. Shapiro, S.S.; Wilk, M.B. An analysis of variance test for normality (complete samples). *Biometrika* **1965**, *52*, 591–611. [\[CrossRef\]](#)
37. R Core Team. *R: A Language and Environment for Statistical Computing*; R Foundation for Statistical Computing: Vienna, Austria, 2023; Available online: <https://www.R-project.org/> (accessed on 5 January 2023).

38. Ravindran, B.; Mupambwa, H.A.; Silwana, S.; Mkeni, P.N. Assessment of nutrient quality, heavy metals and phytotoxic properties of chicken manure on selected commercial vegetable crops. *Heliyon* **2017**, *3*, e00493. [[CrossRef](#)] [[PubMed](#)]
39. Celestina, C.; Hunt, J.R.; Sale, P.W.; Franks, A.E. Attribution of crop yield responses to application of organic amendments: A critical review. *Soil Tillage Res.* **2019**, *186*, 135–145. [[CrossRef](#)]
40. Antonious, G.F.; Chiluwal, A.; Nepal, A. Chitin, Biochar, and Animal Manures Impact on Eggplant and Green Pepper Yield and Quality. *Agric. Sci.* **2023**, *14*, 368–383. [[CrossRef](#)]
41. FAO & WHO. *Food Additives and Contaminants—Joint Codex Alimentarius Commission, FAO/WHO Food standards Program. ALINORM 01/12A*; FAO: Rome, Italy; WHO: Geneva, Switzerland, 2014; p. 1289.
42. Danjuma, M.S.; Abdulkadir, B. Bioaccumulation of heavy metals by leafy vegetables grown with industrial effluents: A review. *Bayero J. Pure Appl. Sci.* **2018**, *11*, 180–185. [[CrossRef](#)]
43. Radulescu, C.; Stihl, C.; Popescu, I.V.; Dulama, I.D.; Chelarescu, E.D.; Chilian, A. Heavy metal accumulation and translocation in different parts of *Brassica oleracea* L. *Rom. J. Phys.* **2013**, *58*, 1337–1354.
44. Smical, A.I.; Hotea, V.; Oros, V.; Juhasz, J.; Pop, E. Studies on transfer and bioaccumulation of heavy metals from soil into lettuce. *Environ. Eng. Manag. J.* **2008**, *7*, 609–615. [[CrossRef](#)]
45. Adriano, D.C.; Adriano, D.C. Other trace elements. In *Trace Elements in the Terrestrial Environment*; Springer: New York, NY, USA, 1986; pp. 470–501. [[CrossRef](#)]
46. Pajević, S.; Arsenov, D.; Nikolić, N.; Borišev, M.; Orčić, D.; Župunski, M.; Mimica-Dukić, N. Heavy metal accumulation in vegetable species and health risk assessment in Serbia. *Environ. Monit. Assess.* **2018**, *190*, 459. [[CrossRef](#)]
47. Ametepey, S.T.; Cobbina, S.J.; Akpabey, F.J.; Duwiejuah, A.B.; Abuntori, Z.N. Health risk assessment and heavy metal contamination levels in vegetables from Tamale Metropolis, Ghana. *Int. J. Food Contam.* **2018**, *5*, 5. [[CrossRef](#)]
48. Arora, M.; Kiran, B.; Rani, S.; Rani, A.; Kaur, B.; Mittal, N. Heavy metal accumulation in vegetables irrigated with water from different sources. *Food Chem.* **2008**, *111*, 811–815. [[CrossRef](#)]
49. Riaz, U.; Aslam, A.; uz Zaman, Q.; Javeid, S.; Gul, R.; Iqbal, S.; Javid, S.; Murtaza, G.; Jamil, M. Cadmium contamination, bioavailability, uptake mechanism and remediation strategies in soil-plant-environment system: A critical review. *Curr. Anal. Chem.* **2021**, *17*, 49–60. [[CrossRef](#)]
50. Tangahu, B.V.; Sheikh Abdullah, S.R.; Basri, H.; Idris, M.; Anuar, N.; Mukhlisin, M. A review on heavy metals (As, Pb, and Hg) uptake by plants through phytoremediation. *Int. J. Chem. Eng.* **2011**, *2011*, 939161. [[CrossRef](#)]
51. Piłkuła, D. Effect of the Degree of Soil Contamination with Cd, Zn, Cu i Zn on Its Content in the Forster Crops and Mobility in the Soil Profile. In *Soil Contamination-Recent Advances and Future Perspectives*; IntechOpen: London, UK, 2023.

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