Quality Assurance and Safety of Crops & Foods, 2025; 17(2): 1–18

Effects of pot simulated heavy metal stress on growth and heavy metal accumulation of *Brassica chinensis L.* in greenhouse

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Academic Editor: Aziz A. Fallah, PhD, Department of Food Hygiene and Quality Control, Faculty of Veterinary Medicine, Shahrekord University, Shahrekord, Iran

Received: 15 March 2024; Accepted: 21 November 2024; Published: 1 April 2025

ORIGINAL ARTICLE

Abstract

The effect of heavy metal migration, accumulation, and growth of *Brassica chinensis L.* were studied by the pot experiment. The results had shown that cadmium (Cd), lead (Pb), copper (Cu), zinc (Zn), and nickel (Ni) had inhibitory effects on the growth of *Brassica chinensis L.*, but chromium (Cr) did not. The highest inhibitory degree was observed for Ni and Cu, relative growth rate was 0.21–0.35 and 0.32–0.42 g d^{-1}, stress tolerance index was 22.44–31.77 and 29.88–36.55%, and leaf area and root length were the lowest. The main accumulation organs of Cd, Cu, and Zn are edible parts, and Pb, Cr, and Ni are in the roots. The bioconcentration factor of Cd (0.212–7.064) is the highest and that of Ni and Cr are the lowest. The accumulation of Cd and Pb, Cd and Zn, and Cd and Ni in *Brassica chinensis L.* showed mutual inhibition effects, and the accumulation of Pb and Cr in *Brassica chinensis L.* showed synergistic effects. In the production of leaf vegetables, attention should be given to the pollution of Cd in soil. *Brassica chinensis L.* has a high cumulative capacity for Cd, which has a certain application prospect in the implementation of green technology remediation of Cd-contaminated soil.

Keywords: red soil; greenhouse; *Brassica chinensis L.*; heavy metals; accumulation

Introduction

Vegetables can provide vitamins, proteins, and other essential nutrients and are one of the main sources of food for humans (Wang *et al.*, 2022). China is a major producer and consumer of vegetables. With the continuous improvement of living standards, vegetable safety has attracted extensive attention. Greenhouse vegetables can balance the supply of vegetables, as it is an efficient production mode. In recent years, with the rapid development of greenhouse vegetables, there have also been problems of environmental pollution and ecological risk, such as a high multiple cropping index, excess pesticides and chemical fertilization, and heavy metals continuously accumulating in greenhouse vegetable fields (Gao *et al.*, 2019; Wang *et al.*, 2021; Wen *et al.*, 2020). Heavy metals cannot be naturally degraded, resulting in their accumulation and pollution (Tariq *et al.*, 2019). Contamination of food by heavy metals has a series of toxic and lethal effects on human health. The major symptoms of heavy metals toxicity in humans are intellectual disability in children, central nervous system disease, dementia and depression in adults, insomnia, kidney and liver diseases, emotional instability, and vision disturbances (Jan *et al.*, 2010). Intake of metals through contaminated vegetables may cause various chronic diseases, such as malnutritional disablities, immunological deficiencies, impaired psychosocial faculties, intrauterine growth retardation, and a high risk of upper gastrointestinal cancer in rats (Türkdogan *et al.*, 2003). Cd can cause diseases such as bronchiolitis, emphysema, and alveolitis. Symptoms of lead poisoning are primarily associated with the central nervous system and gastrointestinal tract, and its poisoning can be either acute or chronic. Zn can cause impairment of growth and reproduction (Nolan, 2003). Cr is toxic to plants and animals including humans (Mohanty *et al.*, 2013). Exposure of vegetables to As can cause abnormal heartbeat, low level of erythrocytes and leukocytes, nausea and vomiting, pricking sensation in the hands and legs, and severe blood vessel damage, cardiovascular disease, hypertension, internal cancers, neurological problems, peripheral vascular disease, pulmonary disease, skin lesions, and diabetes mellitus (Smith *et al.*, 2000). Hg can cause defects such as fetal changes, spontaneous miscarriage, sexual impotency, and low sperm quality. Cu can cause defects such as severe mucosal irritation and corrosion and necrotic changes in the liver and kidney. Ni can affect the pulmonary system and skin, potential carcinogen for lung, and cause skin allergies. Plants absorb heavy metal from the soil in response to concentration gradient and selective uptake of ions or by diffusion. Roots play a significant role in the uptake of heavy metals. The adsorption of heavy metals on the root surface occurs in the cationic form with negative cell wall (Arif *et al.*, 2016). The cations are available at the root surface from the dissociation of their complex forms and accumulated into the apoplast (Krzesłowska, 2011). Once accumulated in the root apoplast, the cations are either retained in the root cells or translocated radially to root stele and subsequently loaded into the xylem and phloem tissues through two ways known as apoplastic passive transportation and symplastic active transportation. The passive transportation occurs through the intercellular spaces by the diffusion of metal ions in the root cell through the soil solution, while the active transportation of heavy metal ions takes place through the plasma membrane mediated by different carriers or transporters (Barberon and Geldner, 2014). The transpiration stream derives the xylem sap upward, where heavy metals are distributed to the part of above ground. Heavy metals are accumulated in photosynthetically active leaves. Many studies have shown that greenhouse vegetable fields in China are polluted by heavy metals. There is a trend of heavy metal accumulation in greenhouse vegetable fields. The migration ability of heavy metals in greenhouse vegetable bases was decreased in the order of Cd>Hg>Cr>As>Pb (Xu *et al.*, 2017). Cd, Cu, and Cr concentrations in greenhouse vegetable fields in the Huang Huai Hai and Bohai Rim regions have exceeded local maximum limit levels (Hu *et al*, 2017). Hu *et al.* (2018) found that Cd concentrations in 17.9% of sampled soils from five greenhouse vegetable production areas exceeded local maximum limit levels, and leafy vegetables have relatively higher concentrations and transfer factors of Cd. Soil under long-term greenhouse planting has seriously accumulated Cd and other heavy metals (Li *et al.*, 2018). The concentrations of Pb and Cr of vegetables in Yunnan, China, have reached two times the region's maximum limit levels (Wang *et al.*, 2021). The concentration of Cd in the greenhouse soil in Xi'an city has exceeded 0.4 mg kg^{-1} (Guo *et al.*, 2022). Liu *et al.* (2021) found that the concentration of Cd in greenhouse soil was 0.84 mg kg^{-1}. The mean concentrations of all eight heavy metals in plastic-shed soils were greater than those in open-field agricultural soils, Zn, Cr, Cd, Hg, and Pb cause heavy pollution in the vegetable fields of the suburb Urumqi in Xinjiang, China (Sawut *et al.*, 2018). Chen *et al.* (2021) found that Cd and As seriously exceeded the maximum limit levels, and the transfer capacity of Cd and Zn in leafy vegetables was the strongest. Yuan *et al.* (2008) and Meng *et al.* (2021) have found that the pollution of Cd, Pb, and Cr in North China greenhouse vegetable fields is relatively serious; transfer factors of As, Cr, Ni, and Pb were higher in leaf vegetables than in tuber and fruit vegetables; and Cd showed relatively higher transfer factors in vegetables than other metals. Leafy vegetables can accumulate higher amount of trace elements (Gupta *et al.*, 2019), and they have a relatively high potential for Cd uptake and translocation (Baldantoni *et al.*, 2016). Yuan *et al.* (2008) used the bioconcentration factor (BCF) to evaluate heavy metal transport ability in greenhouse vegetables in northern China and found that the transport coefficients of As, Cr, and Pb were higher in leafy and fruit vegetables than in tuber vegetables. Study of Gupta *et al.* (2019) showed that the heavy metal transport factor of leafy vegetables was higher. Jalali and Meyari (2022) evaluated the heavy metal transfer factor in the soil–vegetable system, the transfer factor of Cd is the highest, and iron is the lowest. Birghila *et al.* (2023) evaluated the heavy metal transfer factor of tomato; the transfer factor was less than 1. The soil–vegetable system of historical wastewater irrigation in Kaifeng city, central China, was evaluated by BCF, and the transfer factor of Cd was the highest (Cao *et al.*, 2024; Ruan *et al.*, 2023). The heavy metal translocation factor was higher in spinach (Ahmed *et al.*, 2022). Some heavy metals are essential elements for plant growth; however, numerous studies (Chin *et al.*, 2022; Zubair *et al.*, 2024) have shown that when heavy metals reach a certain threshold, they have a significant impact on plant growth, resulting in a reduction in biomass. The Brassica species are known as metal accumulators (Neilson and Rajakaruna, 2012), which have shown high tolerance to heavy metals' concentrations in soils (Mourato *et al.*, 2015). Studies have demonstrated the accumulation of metals in the Brassica species (Purakayastha *et al*., 2008). Brassica chinensis accumulated higher levels of Cd, Cr, and Pb in all plant parts than Brassica rapa (Zunaidi *et al*., 2023). Heavy metals not only stunted plant and crop production growth but also posed a significant threat to ecosystems and food chains. Even at trace levels, plants tend to absorb toxic elements, leading to a growing concern within the community (Ningombam, 2024). There is an urgent need to identify plant species with hyper accumulation capabilities for heavy metals. Simultaneously, further research is needed to explore heavy metals' accumulation, transport, and tolerance in the Brassica species.

Red soil is characteristic of a strong biological enrichment for heavy metals. The soil is polluted by heavy metals, which can easily result in excessive heavy metals in vegetables, especially leafy vegetables (Nie *et al*., 2023; Singh *et al*., 2024; Yvette *et al*., 2024). Therefore, in the case of heavy metal pollution, it is of great significance to explore the migration rule, pollution status, and level of heavy metals in greenhouse vegetables and assess whether vegetable planting activities can continue to ensure the quality and safety of greenhouse vegetables, especially to control heavy metal pollution in leafy vegetables. In this study, greenhouse fields were taken as the research object to simulate soil polluted by Cd, Pb, Cr, Cu, Zn, and Ni at different levels. The purpose of this study was to determine the tolerance and accumulation of six heavy metals, including Cd and Pb, in Brassica plants, which were selected because this vegetable is commonly grown locally and has a high accumulation rate of heavy metals. The objectives of this study were: (1) To observe the growth of brassica under heavy metal pollution; (2) to study the accumulation rate of heavy metals in Brassica and the interaction between heavy metal accumulation; (3) to evaluate its remediation potential for soil heavy metal pollution. This study can provide a reference for the quality and safety assurance of the development of protected vegetables in red soil areas.

Materials and Methods

Experimental materials

The experimental soil is red soil, which was collected from the land (0–20 cm) in the greenhouse of Fuzhou Agricultural Research Institute in Jiangxi Province, China. The physical and chemical properties of the collected red soil are as follows: texture classification: alfisol, sand (0.05–2.00 mm) 30.4%, silt (0.002–0.05 mm) 53.2%, clay (<0.002 mm) 16.4%, electrical conductivity 0.32 dS m^{-1}, moisture 32.5%, total organic carbon 1.82%, organic matter 2.76%, total N 0.14%, total P 0.05%, total K 1.61%, total Ca 1.13%, total Mg 0.85%, total Na 0.11%, cation exchange capacity 13.2 cmol kg^{-1}, total Cd 0.12 mg kg^{-1}, total Pb 37.9 mg kg^{-1}, total Cr 45.9 mg kg^{-1}, total Cu 20.8 mg kg^{-1}, total Zn 49.1 mg kg^{-1}, total Ni 19.3 mg kg^{-1}, and pH 5.42. According to HJ 333-2006, the soil is not polluted. The experimental crop was *Brassica chinensis L.*, produced by the Degao Vegetable Seed Research Institute of Dezhou City, Shangdong Province, China.

Experimental design

The surface soil (0–20 cm, 930 kg) was collected from the vegetable experimental field of Fuzhou Agricultural Science and Technology Institute. The soil was crushed, air-dried, and passed through a 10-mesh nylon sieve. The sifted soil was divided into 93 plastic pots (60×20×15 centimeter), each containing 10 kg of soil. With no heavy metals added as the control and with heavy metals added to the soil reaching the maximum limit levels of HJ 333-2006 as the starting point, in order to simulate the soil by different degrees of heavy metal pollution, heavy metals were added according to Table 1. The amount of added heavy metals was increased according to the gradient to simulate the impact of heavy metals on vegetable growth under different levels of pollution. HJ 333-2006 stipulates that the limited values of Cd, Pb, Cr, Cu, Zn, and Ni are, respectively, 0.3, 50, 150, 50, 200, and 40 mg kg^{-1}. After addition, the content of heavy metals in the soil reached the limit value as the starting point, and then gradually increased according to the multiple of the first addition amount to simulate different soil pollution degrees. At the same time, the pollution degree and migration rule of heavy metals in vegetables were analyzed when the heavy metals exceeded the maximum limit levels. The experimental design is shown in Table 1. Six treatments were set up for fixed planting soil. At the same time, a control group was set up without additives, and each treatment was repeated three times. The heavy metals were applied to the soil in the form of salt solution, fully mixed with the soil, and then sprayed with deionized water to maintain an appropriate amount of water in the soil. After 45 days of balanced cultivation, they will be used as standby.

Table 1. Heavy metal addition scheme.

Addition group	Control group	Heavy metal addition concentration (mg kg^{-1})				
Cd	0	0.2	0.3	0.5	1	2
Pb	0	10	20	30	50	100
Cr	0	100	200	300	400	600
Cu	0	30	50	100	200	400
Zn	0	100	200	300	500	800
Ni	0	20	40	100	200	300

The pot experiment was conducted in the experimental greenhouse of Fuzhou Agricultural Science Research Institute in Jiangxi Province. The pot experiment began at the end of October and ended in December 2022, and the time from sowing to harvesting lasted 65 days.

Sample collection

The soil samples were collected before grinding, mixing, and sieving. Samples were taken from four corner sites around the soil pile and point site in the center, and finally mixed into a single soil sample, and taken back to the lab in a cloth bag. Soil samples were dried at room temperature, finely ground, and passed through a 10-mesh sieve, which was used to determine physicochemical properties. A portion of the soil that was passed through a 100-mesh sieve was used to determine the concentrations of heavy metals. Each pot of *Brassica chinensis L.* was collected as a sample and placed into a ziplock bag when they were ripe and brought back to the laboratory under cold storage conditions. Distilled water was used to wash vegetable samples, and they were dried at room temperature. The *Brassica chinensis L.* samples were divided into leaf, stem, and root parts for weighing. The fresh weight of each part was recorded, in which the leaves and stems were included in the weight of the above ground part, and the root was included in the underground part. Each part was placed into a paper bag for baking for 30 minutes at 100 °C and dried at 70 °C to a constant weight, and then the water content was calculated (Dong *et al.*, 2022; Shuai *et al.*, 2022). The final test results are converted to fresh samples.

Sample analysis and determination method

HMs content of soil and vegetable samples was determined according to the methods by Nie *et al.* (2023) and Chen *et al.* (2021).

The soil analyses included texture classification, pH, cation exchange capacity, organic matter (NY/T 1121-2006), moisture content (NY/T 52-1987), electrical conductivity (HJ 802-2016), total organic carbon content (HJ 658-2013), total N (LY/T 1228-2015), P (LY/T 1232-2015), K (LY/T 1234-2015), Ca, Mg, and Na (NY/T 296-1995).

Data analysis

Edible rate

The edible rate was calculated to assess the edibility of the plants grown in various metal-spiked soil treatments. This calculation involved measuring the total fresh biomass of the species and the fresh biomass of the edible plant parts (leaves and stems). It was calculated as shown in the equation (Eq).1 (Zunaidi *et al.*, 2024).

$$\text{Edible rate} = \frac{\text{fresh biomass of the edible plant parts}}{\text{total fresh biomass of plant}} \times 100\% \quad (1)$$

Relative growth rate

The relative growth rate of a plant was assessed by measuring the plant height under metal-spiked soil treatments over a 65-day period. It was calculated as shown in Equation 2 (Hunt *et al.*, 2002).

$$\text{Relative growth rate} = \frac{\text{final height} - \text{initial height}}{\text{growth day after transplant}} \quad (2)$$

where the final height is the height of the plant on the day of harvesting, and the initial height is the height of the treated plant during the initial measurement. The growth day after transplant is 65 day.

The stress tolerance index

The stress tolerance index was evaluated for plant height by comparing the mean height of treated plants to that of control plants under various heavy metal treatments. It was calculated as shown in Equation 3 (Mi *et al.*, 2019).

$$\text{Stress tolerance index} = \frac{\text{height of treated plant}}{\text{height of control plant}} \times 100\% \quad (3)$$

Leaf area and root length

Leaf areas and root lengths are a crucial aspect in examining how heavy metals affect the growth of vegetables and their ability to adapt in polluted environments. Leaf areas were determined by multiplying the maximum length and width of a leaf. It was calculated as shown in Equation 4 (Zunaidi *et al.*, 2024).

$$\text{Leaf area} = \text{Maximum leaf length} \times \text{Maximum leaf width} \quad (4)$$

The root length was estimated by measuring the maximum length of the main root, from the base to the root tip using a vernier caliper.

BCF

The BCF is used to describe the transfer of heavy metals from the soil to vegetables (Tariq, 2019; Zunaidi *et al.*, 2023). It was calculated as shown in the following equation.

$$\text{BCF} = \frac{C - C_0}{T} \quad (5)$$

where C is the content of heavy metals in the corresponding parts of the addition group, C_0 is the content of heavy metals in the corresponding parts of the control group, and T is the concentration of heavy metals added to the soil.

SPSS 17.0 was used to process raw data and perform statistical analyses of survey results. Part of the figure was drawn by Origin Pro 8.

Results and Discussion

Effects of the addition of heavy metals on the growth of *Brassica chinensis L.*

The effects of different Cd, Pb, Cr, Cu, Zn, and Ni additions on the growth of *Brassica chinensis L.* are shown in Table 2. The effects of the different amounts of added Cd, Pb, Cr, Cu, Zn, and Ni on the growth biomass of *Brassica chinensis L.* are shown in Table 1. The edible rate at each addition level of Cd, Pb, Cr, Cu, Zn, and Ni was 88.66–90.33%, 91.95–93.98%, 94.28–95.86%, 86.68–89.87%, 87.66–96.73%, and 87.34–91.04%, respectively. The relative growth rate at each addition level of Cd, Pb, Cr, Cu, Zn, and Ni was 0.52–0.56, 0.89–1.08, 1.36–1.42, 0.32–0.42, 0.28–1.41, and 0.21–0.35 g d^{-1}, respectively. The stress tolerance index at each addition level of Cd, Pb, Cr, Cu, Zn, and Ni was 43.44–45.84%, 68.34–81.13%, 100.58–104.39%, 29.88–36.55%, 26.64–103.96%, and 22.44–31.77%, respectively. The leaf area at each addition level of Cd, Pb, Cr, Cu, Zn, and Ni was 115.47–127.51, 165.11–212.33, 268.44–281.2, 32.24–49.25, 21.36–282.68, and 19.31–39.7 cm^2. The root length at each addition level of Cd, Pb, Cr, Cu, Zn, and Ni was 15.35–16.23, 17.44–21.62, 27.62–29.16, 14.33–16.62, 14.67–28.66, and 12.27–15.81 cm. Except for the Cr addition group, the biomass of other addition groups was significantly different from that of the control group.

Cd can significantly inhibit the growth of *Brassica chinensis L.*, which is consistent with the results of previous studies (Han *et al*., 2017; Wei, 2019). There was no significant difference in the inhibition degree of different Cd addition levels on the growth of *Brassica chinensis L.*, and it did not increase significantly due to the increase in the added level. Excess Pb has a certain inhibitory effect on the production of *Brassica chinensis L.*, and its inhibition degree presents different degrees of inhibition with increasing addition level. When the addition level was 20 and 100 mg kg^{-1}, the inhibition degree was the largest; when the addition level was 10, 30, and 50 mg kg^{-1}, there was no significant difference in the inhibition degree, and the inhibition degree was far lower than that at addition levels of 20 and 100 mg kg^{-1}. Cr not only has no inhibitory effect on the growth of *Brassica chinensis L.* but also has a certain growth-promoting effect. Different Cr addition levels had no significant effect on the growth of *Brassica chinensis L.*. The result is different from the research results of Dong *et al.* (2022), who believe that *Brassica chinensis L.* is vulnerable to Cr stress. Excess Cu can significantly inhibit the growth of *Brassica chinensis L.* and reduce plant biomass (Naveed *et al*., 2023). The inhibition degree first decreases and then increases, and the inhibition degree is the lowest when 50 mg kg^{-1} Cu is added. Adding a small amount of Zn can promote the growth of *Brassica chinensis L.*, but when adding too much Zn, it will have a significant inhibitory effect on the growth of *Brassica chinensis L.*, and with the increase in the level of addition, the inhibitory effect will continue to increase. The inhibition effect of exogenous Ni on the growth of *Brassica chinensis L.* was the most significant among the six heavy metals, and the inhibition degree basically increased according to the increase in the addition level.

Effects of different Cd additions on heavy metal accumulation in different parts of *Brassica chinensis L.*

The results of the Cd addition are shown in Figure 1. The Cd content in leaves of the control group and from low to high concentrations of heavy metals added was 0.141, 1.413, 1.464, 3.681, 4.339, and 7.664 mg kg^{-1}, respectively. The Cd content in the stems from low to high concentrations of heavy metals added was 0.075, 1.170, 1.129, 1.299, 3.786, and 4.353 mg kg^{-1}, respectively. The Cd content in roots from low to high concentrations of heavy metals added was 0.077, 0.135, 0.141, 0.200, 0.286, and 0.424 mg kg^{-1}, respectively. The BCF of Cd in leaves from low to high concentrations of heavy metals added was 7.064, 4.882, 7.362, 4.339, and 3.832, respectively. The BCF of Cd in the stem from low to high concentrations of heavy metals added was 5.851, 3.764, 2.598, 3.786, and 2.176, respectively. The BCF of Cd in roots from low to high concentrations of heavy metals added was 0.675, 0.471, 0.399, 0.286, and 0.212, respectively.

As shown in Figure 1A, different parts of *Brassica chinensis L.* have different degrees of Cd accumulation. The leaf has the highest degree of Cd accumulation, the root has the lowest degree of Cd accumulation, and the stem has the middle degree of Cd accumulation. This indicates that Cd enters the root system of *Brassica chinensis L.* from the soil but does not accumulate in the root system. After the root system absorbs Cd, it is transported to the stem quickly, and part of the Cd accumulates in the stem, but most of the Cd is transported to the leaf. As a result, a large amount of Cd accumulates in the leaves. The order of the degree of Cd accumulation and migration ability of each part of *Brassica chinensis L.* was leaf > stem > root. With the increase in the Cd content in soil, its migration ability generally declined. The leaf is the main organ of Cd accumulation in *Brassica chinensis L.*. Some studies (Han *et al*., 2017; Wei, 2019) show that the Cd content in the

Table 2. Effects of different amounts of added Cd, Pb, Cr, Cu, Zn, and Ni on the growth biomass of *Brassica chinensis L.*

Different added substances and added concentrations	Number of planted plants in each group (Number of plantings per parallel×Number of parallel)	Above ground fresh weight (g/plant)	Whole plant fresh weight (g/plant)	Edible rate (%)	Relative growth rate (g d^{-1})	Stress tolerance index (%)	Leaf areas (cm^{2})	Root lengths (cm)
Control group	n=6×3	92.18±4.47^{a}	95.62±3.86^{a}	96.75±1.16^{a}	1.36±0.02^{a}	100.00^{a}	260.26±3.56^{a}	24.37±0.94^{a}
Cd (0.2 mg kg^{-1})	n=6×3	39.06±1.04^{b}	43.83±0.97^{b}	89.08±1.07^{b}	0.56±0.01^{b}	45.84±0.25^{b}	121.14±2.11^{d}	15.35±0.86^{c}
Cd (0.3mg kg^{-1})	n=6×3	38.51±1.83^{b}	42.67±1.77^{b}	90.33±1.03^{b}	0.54±0.01^{b}	44.62±0.46^{b}	122.38±1.62^{c}	16.18±0.57^{b}
Cd (0.5mg kg^{-1})	n=6×3	39.27±2.17^{b}	43.91±1.96^{b}	89.42±1.11^{b}	0.56±0.01^{b}	45.92±0.51^{b}	127.51±2.36^{b}	16.23±0.62^{b}
Cd (1.0 mg kg^{-1})	n=6×3	37.54±1.59^{b}	42.03±1.52^{b}	89.28±1.05^{b}	0.53±0.01^{b}	43.96±0.39^{b}	116.22±1.54^{e}	14.51±0.73^{d}
Cd (2.0 mg kg^{-1})	n=6×3	36.82±1.65^{b}	41.54±1.61^{b}	88.66±1.02^{b}	0.52±0.01^{b}	43.44±0.42^{b}	115.47±1.33^{f}	15.37±0.55^{c}
Pb (10mg kg^{-1})	n=6×3	72.94±3.36^{be}	77.58±3.07^{be}	93.98±1.09^{be}	1.08±0.02^{be}	81.13±0.80^{be}	212.33±2.08^{b}	21.62±0.77^{b}
Pb (20mg kg^{-1})	n=6×3	61.83±2.94^{bcdf}	65.92±2.55^{bcdf}	93.74±1.15^{bcdf}	0.90±0.01^{bcdf}	68.94±0.66^{bcdf}	169.61±1.13^{e}	18.71±0.69^{d}
Pb (30 mg kg^{-1})	n=6×3	69.15±1.81^{be}	73.69±1.72^{be}	93.86±1.05^{be}	1.02±0.02^{be}	77.07±0.45^{be}	173.81±1.25^{c}	19.34±0.61^{c}
Pb (50 mg kg^{-1})	n=6×3	70.43±2.24^{be}	75.08±2.19^{be}	93.83±1.02^{be}	1.04±0.02^{be}	78.52±0.57^{be}	171.53±2.04^{d}	18.27±0.54^{d}
Pb (100 mg kg^{-1})	n=6×3	60.11±2.03^{bcdf}	65.35±1.97^{bcdf}	91.95±1.03^{bcdf}	0.89±0.01^{bcdf}	68.34±0.51^{bcdf}	165.11±1.69^{f}	17.44±0.46^{e}
Cr (100 mg kg^{-1})	n=6×3	94.37±4.71^{a}	98.49±4.35^{a}	95.86±1.08^{a}	1.40±0.02^{a}	103.00±1.13^{a}	270.72±3.88^{e}	27.62±1.03^{d}
Cr (200 mg kg^{-1})	n=6×3	91.04±5.09^{a}	96.17±4.76^{a}	94.65±1.07^{a}	1.36±0.02^{a}	100.58±1.23^{a}	268.44±3.16^{f}	27.71±1.28^{d}
Cr (300 mg kg^{-1})	n=6×3	92.29±4.43^{a}	97.25±4.08^{a}	94.98±1.09^{a}	1.38±0.02^{a}	101.70±1.06^{a}	275.69±2.89^{d}	28.46±1.12^{c}
Cr (400 mg kg^{-1})	n=6×3	93.88±6.22^{a}	99.24±5.84^{a}	94.55±1.07^{a}	1.41±0.02 ^{a}	103.79±1.51^{a}	281.20±3.74^{b}	28.55±2.05^{c}
Cr (600 mg kg^{-1})	n=6×3	94.13±5.94^{a}	99.82±5.42^{a}	94.28±1.10^{a}	1.42±0.02^{a}	104.39±1.40^{a}	279.17±2.64^{c}	29.16±1.96^{b}
Cu (30 mg kg^{-1})	n=6×3	28.17±3.16^{bcdef}	31.33±2.94^{bcdef}	89.82±1.07^{bcdef}	0.37±0.01^{bcdef}	32.77±0.76^{bcdef}	45.26±1.05^{c}	15.31±0.65^{c}
Cu (50 mg kg^{-1})	n=6×3	31.44±3.23^{bcd}	34.95±2.88^{bcd}	89.87±1.12^{bcd}	0.42±0.01^{bcd}	36.55±0.75^{bcd}	49.25±1.16^{b}	16.28±0.85^{b}
Cu (100 mg kg^{-1})	n=6×3	30.67±3.07^{bcde}	34.57±2.71^{bcde}	88.63±1.13^{bcde}	0.42±0.01^{bcde}	36.15±0.70^{bcde}	41.34±1.24^{e}	16.62±0.75^{b}
Cu (200 mg kg^{-1})	n=6×3	25.71±2.55^{bdef}	29.64±2.23^{bdef}	86.68±1.14^{bdef}	0.34±0.01^{bdef}	31.00±0.58^{bdef}	42.16±1.67^{d}	14.33±0.45^{d}
Cu (400 mg kg^{-1})	n=6×3	25.02±2.68^{bef}	28.57±2.51^{bef}	87.46±1.07^{bef}	0.32±0.01^{bef}	29.88±0.65^{bef}	32.24±1.52^{f}	15.19±0.66^{c}
Zn (100 mg kg^{-1})	n=6×3	96.01±6.65^{ab}	99.41±6.53^{ab}	96.56±1.02^{ab}	1.41±0.02^{ab}	103.96±1.69^{ab}	282.68±2.44^{b}	28.66±0.95^{b}
Zn (200 mg kg^{-1})	n=6×3	90.49±6.04^{ab}	93.53±5.82^{ab}	96.73±1.04^{ab}	1.32±0.02^{ab}	97.81±1.51^{ab}	264.53±3.08^{c}	28.50±1.08^{b}
Zn (300 mg kg^{-1})	n=6×3	44.08±3.91^{c}	46.72±3.03^{c}	94.25±1.29^{c}	0.60±0.01^{c}	48.86±0.78^{c}	103.19±1.13^{d}	20.25±0.58^{c}
Zn (500 mg kg^{-1})	n=6×3	34.15±2.76^{d}	36.90±2.44^{d}	92.67±1.13^{d}	0.45±0.01^{d}	38.59±0.63^{d}	60.22±1.09^{e}	19.14±0.64^{d}
Zn (800 mg kg^{-1})	n=6×3	22.34±2.51^{e}	25.47±2.38^{e}	87.66±1.05^{e}	0.28±0.01^{e}	26.64±0.62^{e}	21.36±1.17^{f}	14.67±0.33^{e}
Ni (20 mg kg^{-1})	n=6×3	27.54±1.99^{bd}	30.38±1.25^{bd}	90.60±1.59^{bd}	0.35±0.01^{bd}	31.77±0.32^{bd}	39.70±1.21^{b}	15.81±0.41^{b}
Ni (40 mg kg^{-1})	n=6×3	22.08±2.06^{cdef}	24.92±2.11^{cdef}	88.55±0.98^{cdef}	0.27±0.01^{cdef}	26.06±0.55^{cdef}	22.48±1.01^{d}	14.29±0.52^{c}
Ni (100 mg kg^{-1})	n=6×3	25.36±1.82^{bcd}	27.85±1.69^{bcd}	91.04±1.08^{bcd}	0.31±0.01^{bcd}	29.13±0.44^{bcd}	25.21±1.49c	14.35±0.31^{c}
Ni (200 mg kg^{-1})	n=6×3	18.77±1.79^{cef}	21.46±1.85^{cef}	87.38±0.97^{cef}	0.21±0.01^{cef}	22.44±0.48^{cef}	19.31±0.55^{f}	12.27±0.25^{e}
Ni (300 mg kg^{-1})	n=6×3	20.21±2.26^{cef}	23.13±2.14^{cef}	87.34±1.06^{cef}	0.24±0.01^{cef}	24.19±0.55^{cef}	21.64±0.82^{e}	13.34±0.54^{d}

Each value represents the mean of three replicates ± standard error, and the different letters within a column for the same cultivar are significantly different at $p < 0.05$.

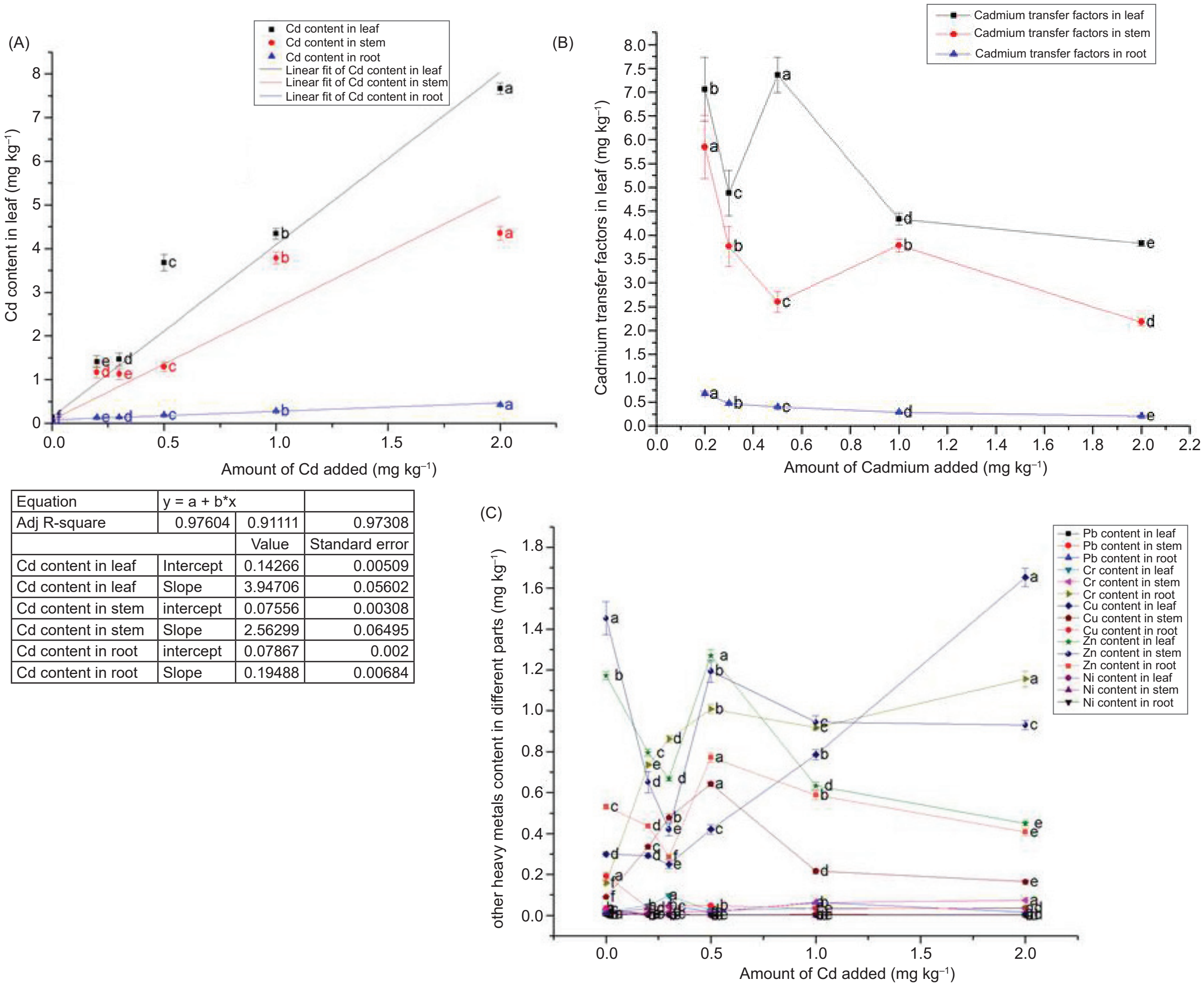
above ground part of *Brassica chinensis L.* is always lower than that in the underground part, which is different from the results of this study. The reason may be that the soil properties are different. The pH and cation exchange capacity of the soil in southern China are lower than those in northern China, leading to a higher proportion of available Cd in the soil and lower ion exchange capacity. The Cd accumulation in each part of *Brassica chinensis L.* was positively correlated with the Cd addition, indicating that the Cd accumulation in each part of *Brassica chinensis L.* increased with the increase in Cd in the soil. When the concentration of added Cd in the leaves of *Brassica chinensis L.* was less than 0.3 mg kg^{-1}, the accumulation capacity of Cd did not change significantly. When the concentration of added Cd reached 0.5 mg kg^{-1}, the accumulation capacity of Cd was greatly enhanced. When the concentration of Cd in the stems of *Brassica chinensis L.* was less than 0.5 mg kg^{-1}, the Cd accumulation capacity did not change significantly. When the concentration of Cd was more than 0.5 mg kg^{-1}, the Cd accumulation capacity was also greatly enhanced. The Cd accumulation ability of roots did not increase significantly with increasing exogenous concentration. Many studies (Ahmed *et al.*, 2022; Cao *et al.*, 2024; Huang *et al.*, 2020; Kalkhajeh *et al.*, 2020) show that leafy vegetables have a strong Cd accumulation capacity. This test shows that after adding exogenous Cd, the Cd content in the leaves and stems of *Brassica chinensis L.* exceeds 1 mg kg^{-1}, seriously exceeding the limit value (0.2 mg kg^{-1}) specified in the national food safety standard GB2762-2017. The comparison between Table 1 and Figure 1A shows that the growth inhibition degree of *Brassica chinensis L.* does not change significantly with the increase in Cd accumulation in each part. Figure 1B shows that the order of the migration coefficient of Cd in various parts of *Brassica chinensis L.* is leaf>stem>root. This is different from the observation by Zunaidi *et al.* (2024) that the distribution of Cd in *Brassica chinensis L.* is root>leaf>stem. With increasing concentration, the migration coefficient of Cd in all parts of *Brassica chinensis L.* was the highest at an addition amount of 0.5 mg kg^{-1}, and the overall migration coefficient of Cd in all parts of *Brassica chinensis L.* gradually decreased. The effect of different amounts of Cd additions on the accumulation of other heavy metals is shown in Figure 1C. The accumulation of Pb and Ni, Cr in leaves and stems, and Cu in roots was not obvious but significantly affected the accumulation of Cr in roots, Cu in leaves and stems, and Zn in all parts. With the increase in Cd content, the accumulation of Cr in roots and Cu in leaves also increased correspondingly. The accumulation of Cu in stems first increased and then decreased, while the accumulation of Zn in all parts of *Brassica chinensis L.* showed a trend of first decreasing, then increasing, and then decreasing. The bioaccumulation coefficient of Cd in vegetables is higher than that of other heavy metals, which may be closely related to Cd bioavailability in the environment, soil pH, and gene expression in plant cells (Lin *et al.*, 2024).

Effects of different Pb additions on heavy metal accumulation

Effects of different Pb additions on heavy metal accumulation in different parts of *Brassica chinensis L.* The different letters within a column for the same cultivar are significantly different at $p < 0.05$.

The results of the Pb addition are shown in Figure 2. The Pb content in leaves from low to high concentrations of heavy metals added was 0.029, 0.050, 0.062, 0.130, 0.118, and 0.167 mg kg^{-1}, respectively. The Pb content in the stems from low to high concentrations of heavy metals added was 0.036, 0.112, 0.348, 0.641, 0.531, and 0.359 mg kg^{-1}, respectively. The Pb content in roots from low to high concentrations of heavy metals added was 0.011, 0.503, 1.892, 2.450, 4.073, and 3.957 mg kg^{-1}, respectively. The BCF of Pb in leaves from low to high concentrations of heavy metals added was 0.005, 0.003, 0.004, 0.002, and 0.002, respectively. The BCF of Pb in the stem from low to high concentrations of heavy metals added was 0.011, 0.017, 0.021, 0.011, and 0.004, respectively. The BCF of Pb in roots from low to high concentrations of heavy metals added was 0.050, 0.095, 0.083, 0.082, and 0.040, respectively.

Figure 2A shows that different parts of *Brassica chinensis L.* have different levels of Pb accumulation. In addition, the accumulation in roots was much higher than that in leaves and stems, the accumulation in stems was in the middle, and the accumulation in leaves was the lowest. Some studies (Zunaidi *et al.*, 2023) show that the Pb content in the above ground part of *Brassica chinensis L.* is higher than that in the underground part, which is different from the results of this study. This shows that roots are the main organ for Pb accumulation in *Brassica chinensis L.*, and the accumulation of Pb in stems and leaves is much lower than that in roots. This finding is consistent with Mi *et al.* (2024). When the level of Pb addition was close to 50 mg kg^{-1}, the accumulation of Pb in roots and stems showed a downward trend. When the concentration of added Pb reached 20 mg kg^{-1}, the Pb content in the stems of *Brassica chinensis L.* was higher than 0.3 mg kg^{-1} (the limit value in GB2762-2017). When the concentration of added Pb reached 30 mg kg^{-1}, the Pb accumulation in the stems of *Brassica chinensis L.* reached the highest level. When the concentration of Pb addition was 30–70 mg kg^{-1}, the Pb accumulation in the stems was more than 0.5 mg kg^{-1}, while the cumulative amount in the leaves did not exceed 0.2 mg kg^{-1}. Figure 2B shows that the transfer coefficient of Pb in different parts of *Brassica chinensis L.* was in the order of root > stem > leaf. This is different from the observation by Zunaidi *et al.* (2024)



Equation	y = a + b*x		
Adj R-square	0.97604	0.91111	0.97308
		Value	Standard error
Cd content in leaf	Intercept	0.14266	0.00509
Cd content in leaf	Slope	3.94706	0.05602
Cd content in stem	intercept	0.07556	0.00308
Cd content in stem	Slope	2.56299	0.06495
Cd content in root	intercept	0.07867	0.002
Cd content in root	Slope	0.19488	0.00684

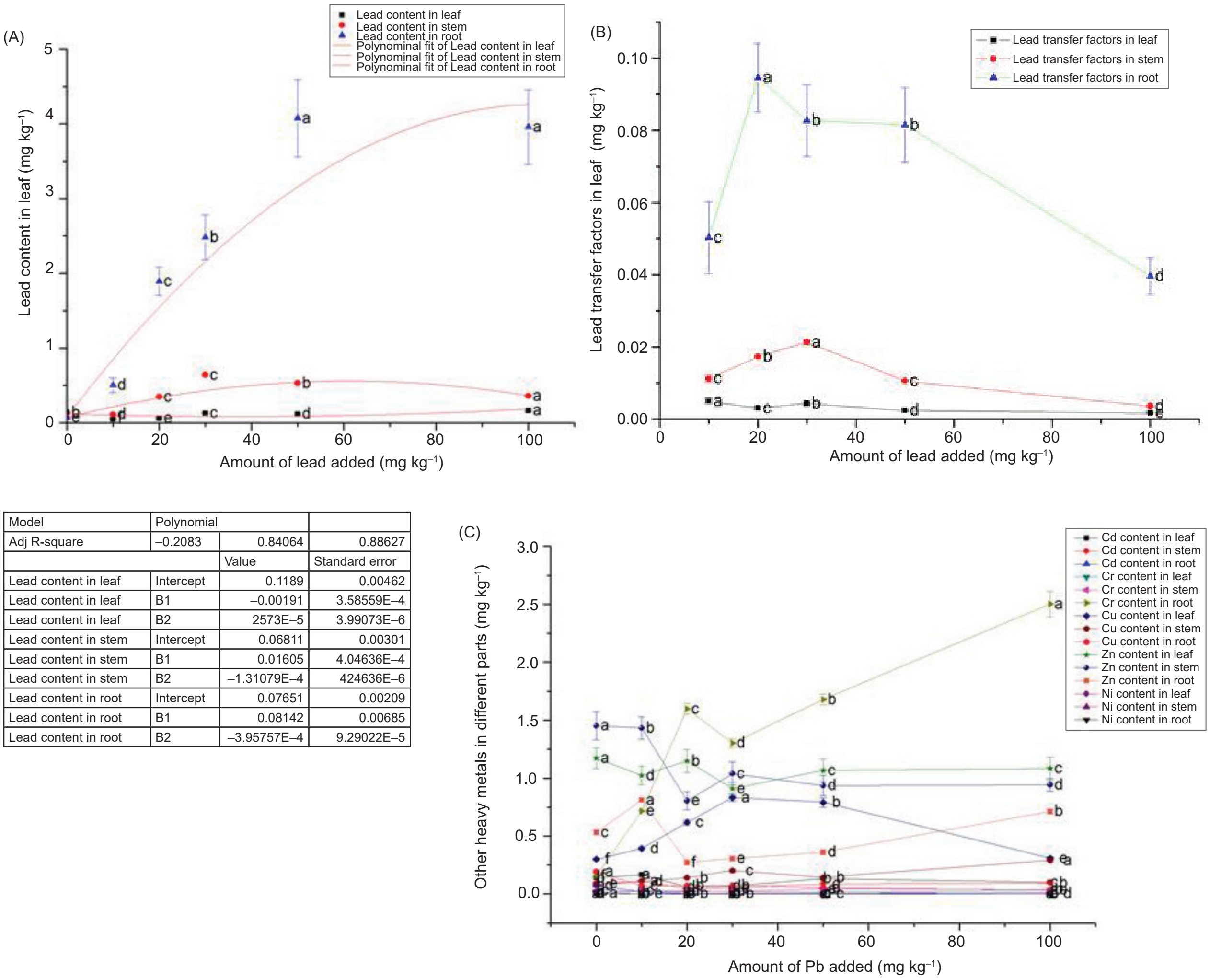
Figure 1. Effects of different concentrations of exogenous Cd on heavy metal accumulation in *Brassica chinensis L.* (A) Cd accumulation of different parts, (B) BCF of Cd in different parts, and (C) accumulation of other heavy metals in different parts.

that the distribution of Cd in *Brassica chinensis L.* is root > leaf > stem, and the transfer coefficient of Pb in roots was much higher than that in stems and leaves. When the level of Pb addition was 20 mg kg^{-1}, the migration coefficient of roots to Pb reached the highest level, and with the increase in the level of Pb addition, the migration coefficient of roots to Pb decreased gradually. The migration coefficient of the stem to Pb showed a trend of first increasing and then decreasing. When the addition level was 30 mg kg^{-1}, the migration coefficient reached the maximum. The migration coefficient of leaves showed a decreasing trend as the concentration increased. The effect of different amounts of Pb additions on the accumulation of other heavy metals is shown in Figure 2C. With the increase in Pb content, the accumulation capacity of Cd and Zn in each part of the plant was inhibited, and the accumulation capacity of Cu in the root was also inhibited. However, the accumulation capacity of Cr in the root was greatly enhanced, while the accumulation capacity of Cu in the leaves and stems showed a trend of first increasing and then decreasing.

Effects of different Cr additions on heavy metal accumulation

Effects of different Cr additions on heavy metal accumulation in different parts of *Brassica chinensis L.* The different letters within a column for the same cultivar are significantly different at $p < 0.05$.

The results of Cr addition are shown in Figure 3. The Cr content in leaves from the control group and from low to high concentrations of heavy metals added was 0.018, 0.026, 0.044, 0.071, 0.143, and 0.224 mg kg^{-1}, respectively. The Cr content in the stems from low to high concentrations of heavy metals added was 0.029, 0.038, 0.059, 0.090, 0.200, and 0.240 mg kg^{-1}, respectively. The Cr

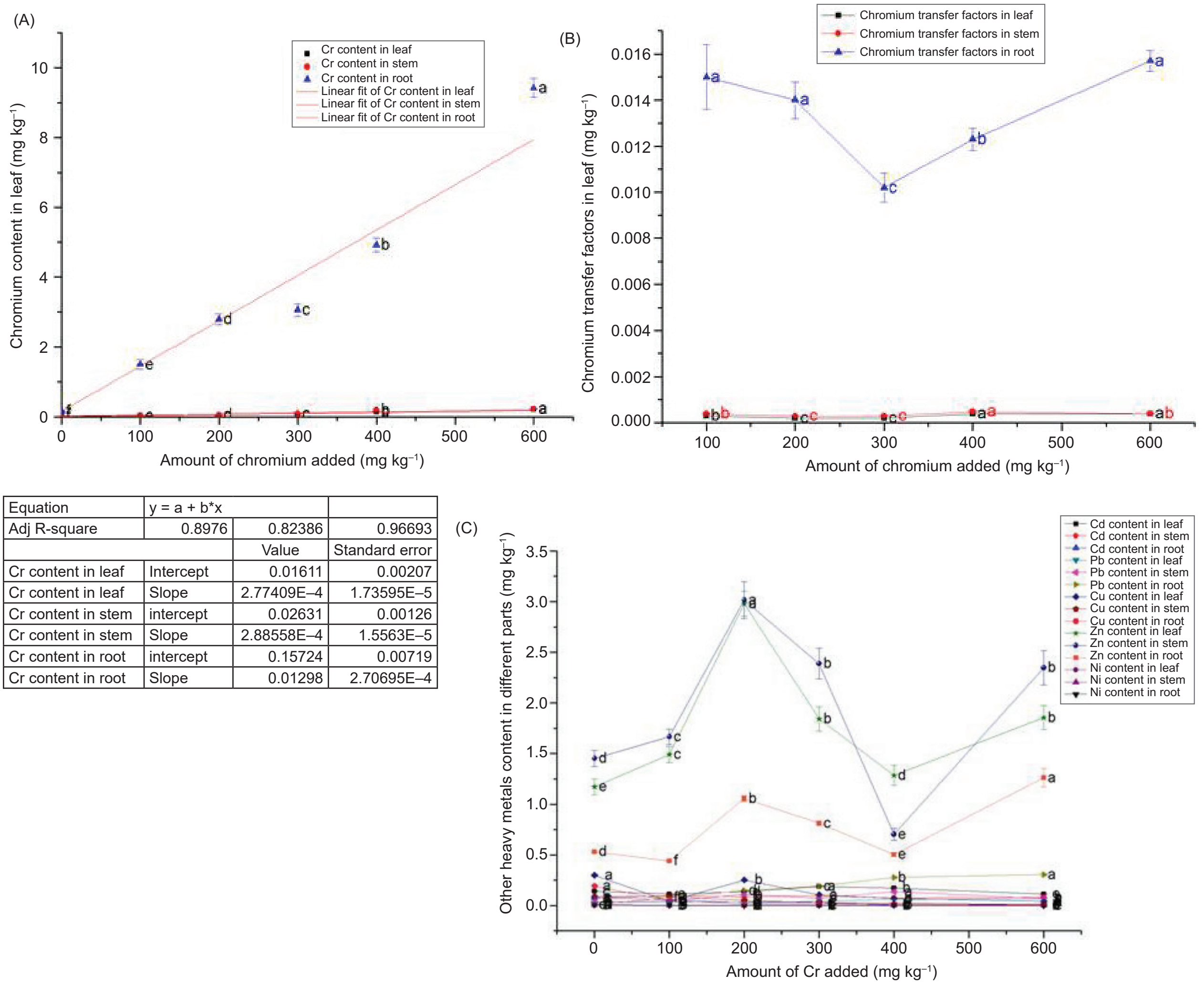


Model	Polynomial		
Adj R-square	–0.2083	0.84064	0.88627
		Value	Standard error
Lead content in leaf	Intercept	0.1189	0.00462
Lead content in leaf	B1	–0.00191	3.58559E–4
Lead content in leaf	B2	2573E–5	3.99073E–6
Lead content in stem	Intercept	0.06811	0.00301
Lead content in stem	B1	0.01605	4.04636E–4
Lead content in stem	B2	–1.31079E–4	424636E–6
Lead content in root	Intercept	0.07651	0.00209
Lead content in root	B1	0.08142	0.00685
Lead content in root	B2	–3.95757E–4	9.29022E–5

Figure 2. Effects of different concentrations of exogenous Pb on heavy metal accumulation in *Brassica chinensis L.* (A) Pb accumulation of different parts, (B) BCF of Pb in different parts, and (C) accumulation of other heavy metals in different parts.

content in roots from low to high concentrations of heavy metals added was 0.158, 1.503, 2.793, 3.061, 4.924, and 9.416 mg kg^{-1}, respectively. The BCF of Cr in leaves from low to high concentrations of heavy metals added was 0.0003, 0.0002, 0.0002, 0.0004, and 0.0004, respectively. The BCF of Cr in stems from low to high concentrations of heavy metals added was 0.0004, 0.0003, 0.0003, 0.0005, and 0.0004, respectively. The BCF of Cr in roots from low to high concentrations of heavy metals added was 0.015, 0.014, 0.010, 0.012, and 0.016, respectively.

Figure 3A shows that within the range of added concentrations, the Cr content in each part of *Brassica chinensis L.* was positively correlated with the Cr content in the soil. With the increase in the Cr content in soil, the Cr accumulation in the roots of *Brassica chinensis L.* increased significantly, while the accumulation in the leaves and stems did not show a significant upward trend with the increase in exogenous Cr concentration. The accumulative amount of Cr in leaves and stems was equivalent and lower than that in roots, which could be attributed to the sequestration of Cr in the vacuoles of root cells as a protective mechanism, thereby naturally enhancing the tolerance of the two species to Cr toxicity (Liu, 2010; Zunaidi *et al.*, 2024). This is consistent with the conclusion that the accumulation of Cr in *Brassica chinensis L.* occurs mainly in roots (Liu, 2010; Yu *et al.*, 2023; Zunaidi *et al.*, 2023). Within the range of addition, the accumulative amount of Cr does not exceed 0.25 mg kg^{-1}, which is far below the limit of 0.5 mg kg^{-1} in the local food safety standard GB2762-2017. Figure 3B shows that the transfer coefficient of Cr in various parts of *Brassica chinensis L.* was in the order of root > stem > leaf, and the transfer coefficient of Cr in roots was much higher than that in stems and leaves. The removal coefficient of Cr in stems and leaves was similar, but the change was not significant with increasing Cr concentration. The effect of different amounts of Pb additions on the accumulation



Equation	y = a + b*x		
Adj R-square	0.8976	0.82386	0.96693
		Value	Standard error
Cr content in leaf	Intercept	0.01611	0.00207
Cr content in leaf	Slope	2.77409E–4	1.73595E–5
Cr content in stem	intercept	0.02631	0.00126
Cr content in stem	Slope	2.88558E–4	1.5563E–5
Cr content in root	intercept	0.15724	0.00719
Cr content in root	Slope	0.01298	2.70695E–4

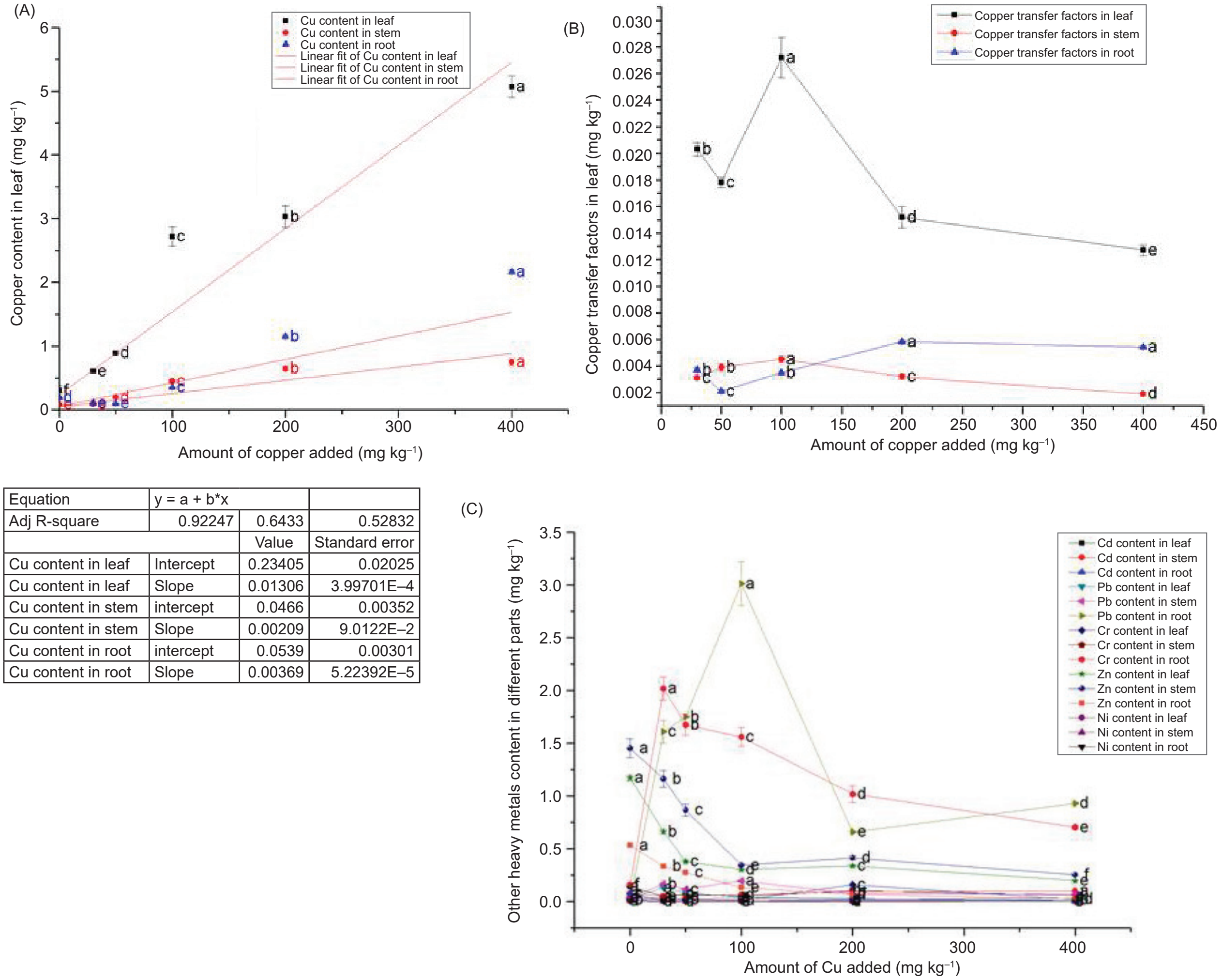
Figure 3. Effects of different concentrations of exogenous Cr on heavy metal accumulation in *Brassica chinensis L.* (A) Cr accumulation of different parts, (B) BCF of Cr in different parts, and (C) accumulation of other heavy metals in different parts. The different letters within a column for the same cultivar are significantly different at p < 0.05.

of other heavy metals is shown in Figure 3C. With the increase in Cr addition, the accumulation of Pb in leaves and roots and Zn in each part showed an increasing trend; the accumulation of Cd and Cu in leaves and Pb in stems first increased and then decreased; and the accumulation of Cd and Pb in roots, Cu in stems, and Ni in each part showed a decreasing trend.

Effects of different Cu additions on heavy metal accumulation in different parts of *Brassica chinensis* L.

The results of Cu addition are shown in Figure 4. The Cu contents in leaves from low to high concentrations of heavy metals added were 0.300, 0.608, 0.890, 2.719, 3.030, and 5.070 mg kg^{-1}, respectively. The Cu content in the stems from low to high concentrations of heavy metals added was 0.090, 0.092, 0.197, 0.450, 0.646, and 0.751 mg kg^{-1}, respectively. The Cu content in roots from low to high concentrations of heavy metals added was 0.193, 0.111, 0.103, 0.351, 1.153, and 2.167 mg kg^{-1}, respectively. The BCF of Cu in leaves from low to high concentrations of heavy metals added was 0.020, 0.018, 0.027, 0.015, and 0.013, respectively. The BCF of Cu in the stem from low to high concentrations of heavy metals added was 0.003, 0.004, 0.005, 0.003, and 0.002, respectively. The BCF of Cu in roots from low to high concentrations of heavy metals added was 0.004, 0.002, 0.004, 0.006, and 0.005, respectively.

Cu plays a vital role in many physiological activities, such as photosynthesis, respiration, and the regulation of the antioxidant system of plants, as well as in the growth and development of plants. However, Cu is considered to be one of the most toxic heavy metals. When plants are exposed to very high concentrations of Cu, it will Pb to the disorder of plant physiological functions and growth retardation (Kumar *et al.*, 2021). Figure 4



Equation	y = a + b*x		
Adj R-square	0.92247	0.6433	0.52832
		Value	Standard error
Cu content in leaf	Intercept	0.23405	0.02025
Cu content in leaf	Slope	0.01306	3.99701E–4
Cu content in stem	intercept	0.0466	0.00352
Cu content in stem	Slope	0.00209	9.0122E–2
Cu content in root	intercept	0.0539	0.00301
Cu content in root	Slope	0.00369	5.22392E–5

Figure 4. Effects of different concentrations of exogenous Cu on heavy metal accumulation in *Brassica chinensis L.* (A) Cu accumulation of different parts, (B) BCF of Cu in different parts, and (C) accumulation of other heavy metals in different parts.

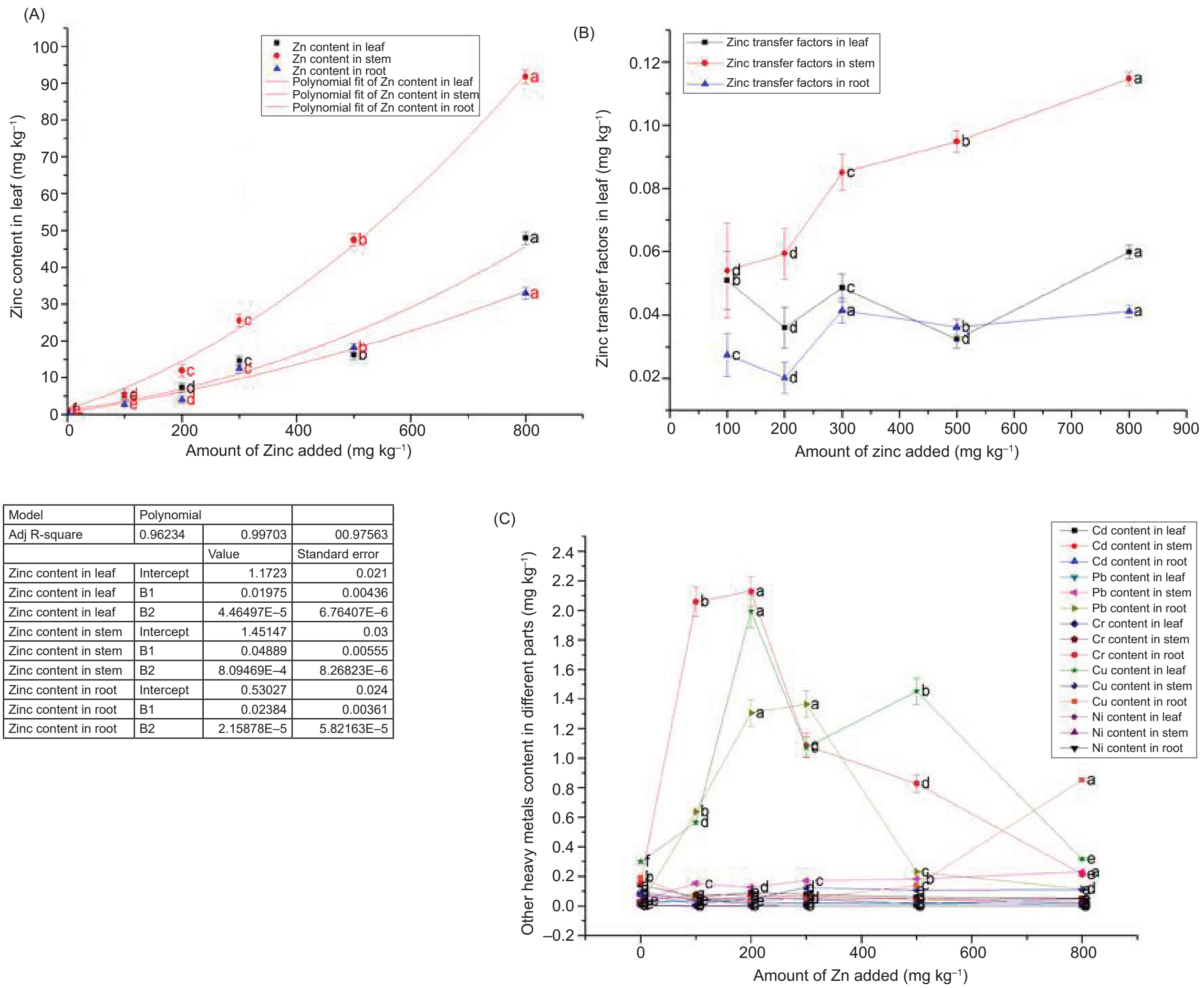
shows that within the range of addition concentrations, the Cu content in *Brassica chinensis L.* is positively correlated with the Cu content added to the soil, but the Cu accumulation effect in each part of *Brassica chinensis L.* is different. The Cu accumulation ability in the leaf part is much higher than that in the stem and root. Zunaidi *et al.* (2023) concluded that the roots of *Brassica chinensis L.* had a stronger Cu accumulation capacity than the leaves and stems, which was different from the results of this study. The Cu accumulation ability in the stem part was the lowest, and the Cu accumulation amount in the leaf part was much higher than that in other parts. The reason is that the leaf is the main organ for photosynthesis, and photosynthesis requires the participation of Cu. It can be seen from the curve trend in Figure 4A that the cumulative amount of Cu in leaves and stems will show a downward trend after reaching a certain degree. With increasing Cu concentration in the environment, the Cu accumulation rate in the roots of *Brassica chinensis L.* increased steadily, and its accumulation capacity exceeded that in the leaves and stems. This result is consistent with the research results of Yuan *et al.* (2008). Figure 4B shows that the migration coefficient of Cu at the leaf position is the highest. With the continuous increase in Cu addition concentration, the migration coefficient of the leaf presents a trend of first decreasing, then increasing, and then decreasing. At a concentration of 100 mg kg^{-1}, the migration coefficient reaches the highest value. The Cu transport coefficient of stems and roots was much lower than that of leaves. The effect of different amounts of Pb additions on the accumulation of other heavy metals is shown in Figure 4C. With the increase in Cu content, the accumulation of Cd in the stem and Pb and Cr in the root showed an increasing trend, the accumulation of Pb in the leaf and stem showed an increasing trend and then a decreasing trend, and the accumulation of Cd in the leaf and root and Zn and Ni in each part showed a decreasing trend.

Effects of different Zn additions on heavy metal accumulation

Effects of different Zn additions on heavy metal accumulation in different parts of *Brassica chinensis L.*. The different letters within a column for the same cultivar are significantly different at $p < 0.05$.

The results of the Zn addition are shown in Figure 5. The Zn content in leaves from the control group and from low to high concentrations of heavy metals added was 1.172, 5.093, 7.209, 14.570, 16.202, and 47.911 mg kg^{-1}, respectively. The Zn contents in the stems from low to high concentrations of heavy metals added were 1.452, 5.401, 11.888, 25.510, 47.407, and 91.721 mg kg^{-1}, respectively. The Zn content in roots from low to high concentrations of heavy metals added was 0.531, 2.741, 4.012, 12.415, 18.088, and 32.928 mg kg^{-1}, respectively. The BCF of Zn in leaves from low to high concentrations of heavy metals added was 0.051, 0.036, 0.049, 0.032, and 0.060, respectively. The BCF of Zn in the stems from low to high concentrations of heavy metals added was 0.054, 0.059, 0.085, 0.095, and 0.115, respectively. The BCF of Zn in roots from low to high concentrations of heavy metals added was 0.027, 0.020, 0.041, 0.036, and 0.041, respectively.

Figure 5A shows that within the range of added concentrations, the Zn content in *Brassica chinensis L.* is positively correlated with the Zn content in the soil. Among them, the stem has the strongest cumulative capacity, and the root has the lowest cumulative capacity. When the Zn addition concentration in the soil reached 300 mg kg^{-1}, the cumulative capacity of the stem and leaf to Zn was enhanced, but the cumulative capacity of the root to Zn did not show rapid growth. An appropriate amount of Zn application can alleviate the toxicity of Cd and other heavy metals and promote plant growth. When Zn



Model	Polynomial		
Adj R-square	0.96234	0.99703	00.97563
		Value	Standard error
Zinc content in leaf	Intercept	1.1723	0.021
Zinc content in leaf	B1	0.01975	0.00436
Zinc content in leaf	B2	4.46497E–5	6.76407E–6
Zinc content in stem	Intercept	1.45147	0.03
Zinc content in stem	B1	0.04889	0.00555
Zinc content in stem	B2	8.09469E–4	8.26823E–6
Zinc content in root	Intercept	0.53027	0.024
Zinc content in root	B1	0.02384	0.00361
Zinc content in root	B2	2.15878E–5	5.82163E–5

Figure 5. Effects of different concentrations of exogenous Zn on heavy metal accumulation in *Brassica chinensis L.* (A) Zn accumulation of different parts, (B) BCF of Zn in different parts, and (C) accumulation of other heavy metals in different parts.

is applied excessively, the ability to mitigate the toxicity of other heavy metals decreases significantly, which in turn inhibits plant growth, thereby decreasing the above ground biomass of plants (Shuai *et al.*, 2022). Combined with the results in Table 1, when the Zn accumulation in *Brassica chinensis L.* increased by 10 mg kg^{-1}, Zn did not play a role in toxicity mitigation but inhibited the growth of *Brassica chinensis L.*. The higher the Zn accumulation in *Brassica chinensis L.* was, the higher the growth inhibition degree of *Brassica chinensis L.*. At the same time, Figure 5B shows that the migration coefficient of each part of *Brassica chinensis L.* is stem > leaf > root, and the migration coefficient of Zn from the stem increases with increasing Zn concentration. The effect of different amounts of Pb additions on the accumulation of other heavy metals is shown in Figure 5C. With the increase in Zn addition, the accumulation of Cd and Ni in all parts showed an inhibitory effect, the accumulation of Pb in stems showed a synergistic effect, the accumulation of Pb, Cr, and Cu in leaves showed a trend of first increasing and then decreasing, the accumulation of Cr in stems showed a trend of first increasing and then decreasing, and the accumulation of Pb and Cr in roots also showed a trend of first increasing and then decreasing. The accumulation of Cu in stems and roots was inhibited first and then synergistically.

Effects of different Ni additions on heavy metal accumulation

Effects of different Ni additions on heavy metal accumulation in different parts of *Brassica chinensis L.*. The different letters within a column for the same cultivar are significantly different at $p < 0.05$.

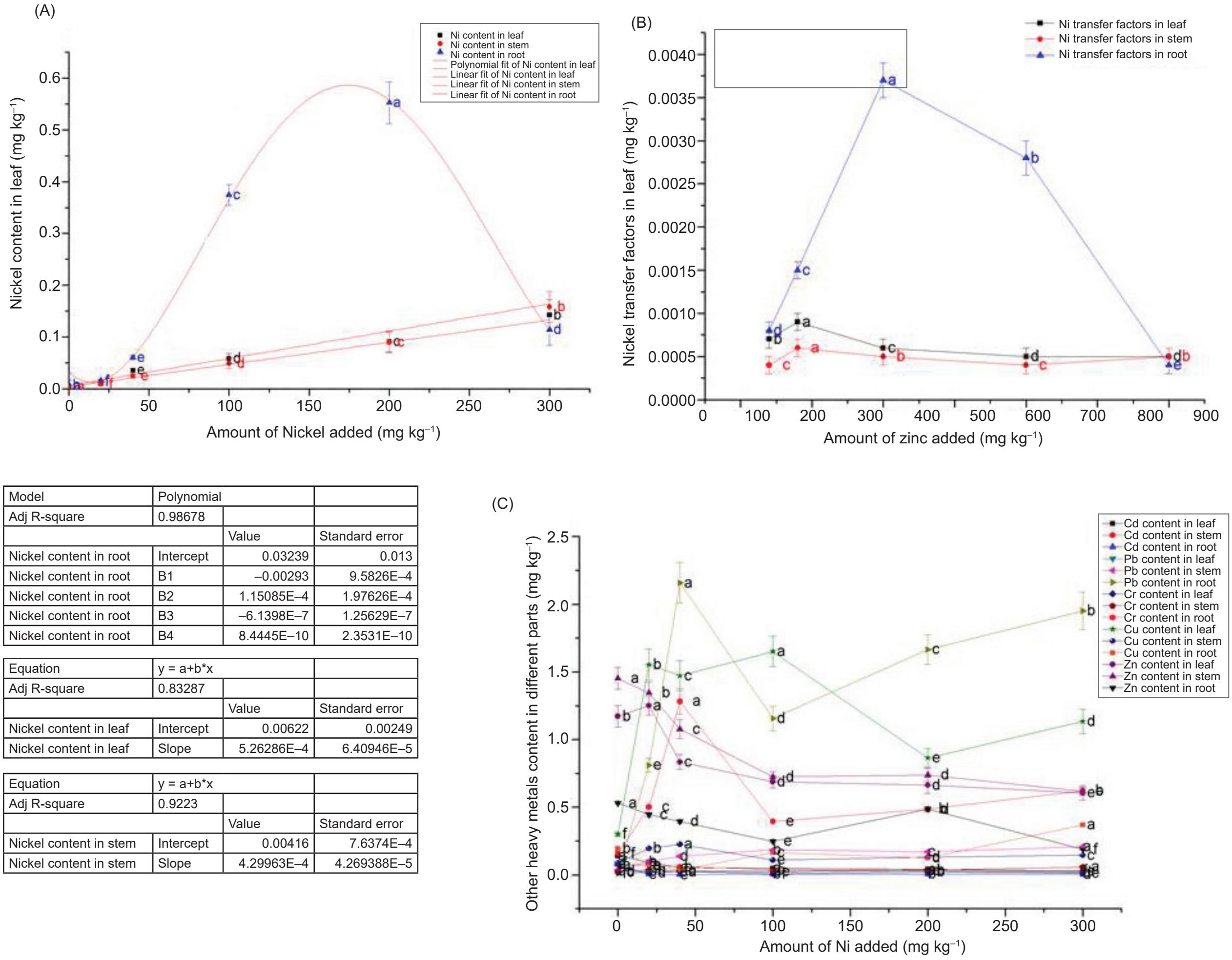
The results of the Ni addition are shown in Figure 3. The Ni content in leaves from low to high concentrations of heavy metals added was 0.006, 0.014, 0.036, 0.059, 0.091, and 0.142 mg kg^{-1}, respectively. The Ni content in the stems from low to high concentrations of heavy metals added was 0.005, 0.009, 0.025, 0.049, 0.090, and 0.158 mg kg^{-1}, respectively. The Ni content in roots from low to high concentrations of heavy metals added was 0.007, 0.016, 0.061, 0.374, 0.552, and 0.114 mg kg^{-1}, respectively. The BCF of Ni in leaves from low to high concentrations of heavy metals added was 0.0007, 0.0009, 0.0006, 0.0005, and 0.0005, respectively. The BCF of Ni in the stem from low to high concentrations of heavy metals added was 0.0004, 0.0006, 0.0005, 0.0004, and 0.0005, respectively. The BCF of Ni in roots from low to high concentrations of heavy metals added was 0.0008, 0.0015, 0.0037, 0.0028, and 0.0004, respectively.

Figure 6A shows that within the range of addition concentrations, the Ni accumulation capacity of the leaves and stems of the above ground part of *Brassica chinensis L.* was positively correlated with the Ni content in the soil, and there was no significant difference in the Ni accumulation capacity of the leaves and stems. When the addition concentration was less than 200 mg kg^{-1}, the Ni accumulation capacity of the leaves and stems did not exceed 0.1 mg kg^{-1}, but the accumulation capacity of the stems was lower than that of the leaves at a lower concentration, and at a higher concentration, Ni accumulation in stems was higher than that in leaves. The Ni accumulation capacity of roots first increased and then decreased with increasing concentration. When the amount of Ni added reached 200 mg kg^{-1}, the Ni accumulation capacity of roots reached a maximum and then showed a downward trend. Within the range of experimental additions, the Ni accumulation organ was mainly roots. Some studies have shown that some vegetables are highly sensitive to Ni. When the accumulation of Ni in plants is greater, the toxicity of Ni is stronger, and the degree of inhibition of vegetable growth is higher (Bai *et al.*, 2018). Combined with the results in Table 1, these results show that *Brassica chinensis L.* is highly sensitive to Ni, and the higher the Ni accumulation in vegetables, the stronger is the growth inhibition. Compared with other heavy metals, Ni has the lowest migration capacity, which is consistent with the findings of Singh *et al.* (2024). Figure 6B shows that the order of the Ni transfer coefficient in each part of *Brassica chinensis L.* is root > leaf > stem. With increasing Ni concentration, the Ni transfer coefficient in each part of *Brassica chinensis L.* first increased and then decreased. When the Ni concentration reaches 100 mg kg^{-1}, the Ni transfer coefficient in roots reaches the highest value. The effect of different amounts of Pb additions on the accumulation of other heavy metals is shown in Figure 6C. With increasing Ni content, the stem and root had a synergistic effect on Pb and Cr accumulation, and the leaves and stem had an inhibitory effect on Cd and Zn accumulation.

There are still some shortcomings in this study; for example, only a single vegetable variety was studied, but there are differences in the corresponding and cumulative ability of heavy metal stress among different varieties. At the same time, multiple heavy metal combined pollution is often encountered in reality, and this study only simulates the situation of a single heavy metal pollution. There is a lack of comparative studies on the effects of heavy metal pollution on crop growth and heavy metal accumulation under different production modes in greenhouses and open fields.

Conclusions

Excessive Cd, Pb, Cu, Zn, and Ni all have inhibitory effects on the growth of *Brassica chinensis L.*, with Ni



Model	Polynomial		
Adj R-square	0.98678		
		Value	Standard error
Nickel content in root	Intercept	0.03239	0.013
Nickel content in root	B1	–0.00293	9.5826E–4
Nickel content in root	B2	1.15085E–4	1.97626E–4
Nickel content in root	B3	–6.1398E–7	1.25629E–7
Nickel content in root	B4	8.4445E–10	2.3531E–10

Equation	y = a+b*x		
Adj R-square	0.83287		
		Value	Standard error
Nickel content in leaf	Intercept	0.00622	0.00249
Nickel content in leaf	Slope	5.26286E–4	6.40946E–5

Equation	y = a+b*x		
Adj R-square	0.9223		
		Value	Standard error
Nickel content in stem	Intercept	0.00416	7.6374E–4
Nickel content in stem	Slope	4.29963E–4	4.269388E–5

Figure 6. Effects of different concentrations of exogenous Ni on heavy metal accumulation in *Brassica chinensis L.* (A) Ni accumulation of different parts, (B) BCF of Ni in different parts, and (C) accumulation of other heavy metals in different parts. The different letters within a column for the same cultivar are significantly different at $p < 0.05$.

and Cu having the highest inhibitory degree, followed by Cd. Cr had no inhibitory effect on the growth of *Brassica chinensis L.*. The main accumulation organs of Cd, Cu, and Zn are the edible parts of stems and leaves, and the main accumulation organs of Pb, Cr, and Ni are mainly in the roots. The BCF of Cd is much higher than that of the five heavy metals, and the BCF of Ni and Cr are the lowest. Compared with other heavy metals, *Brassica chinensis L.* has a high cumulative capacity for Cd, which has a certain application prospect in the implementation of green technology remediation of Cd-contaminated soil. The accumulation of Cd and Pb, Cd and Zn, and Cd and Ni in *Brassica chinensis L.* showed mutual inhibition effects, and the accumulation of Pb and Cr in *Brassica chinensis L.* showed synergistic effects. In the production of leaf vegetables, attention should be given to the pollution of Cd in soil. In the future, it is necessary to further study the accumulation and growth of heavy metals in vegetables under different vegetable varieties, heavy metal composite pollution, and different production modes.

Statements and Declarations

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgment

This study was financially supported by the Jiangxi Vegetable Industry Technical System (Grant No.: JXARS-06).

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Author Contributions

Genxin Nie was in charge of conceptualization, methodology, investigation, editing, writing, and original draft. Yaomin Zhou was responsible for resources, supervision, and review & editing. Tianhua Tu did analysis and data collection. Lifang Hu performed sample collection. Ling Wu was concerned with methodology and review & editing. All authors have read and agreed to the published version of the manuscript.

Funding

This work had received a grant from the Jiangxi Provincial Department of Agriculture and Rural Affairs, China.

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