

<https://doi.org/10.48047/AFJBS.7.4.2025.923-935>



African Journal of Biological Sciences

Journal homepage: <http://www.afjbs.com>



Research Paper

Open Access

Effects of Lead Toxicity on Iron and Zinc Accumulation in Four Evergreen Plant Species: A Controlled Pot Experiment

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Volume: 7, Issue 4, Apr 2025

Received: 15 Feb 2025

Accepted: 05 Mar 2025

Published: 05 Apr 2025

[doi:10.48047/AFJBS.7.4.2025.923-935](https://doi.org/10.48047/AFJBS.7.4.2025.923-935)

Abstract:

Background and aim: Lead (Pb) toxicity poses environmental challenges, disrupting plant growth and micronutrient dynamics. This study investigated the impact of Pb toxicity on iron (Fe) and zinc (Zn) accumulation in four evergreen species: *Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benjamina*.

Method: A pot experiment was conducted in Erbil, Iraq, using sandy clay loam soil treated with Pb at 0, 100, 200, and 300 mg/kg. Plants were grown for six months in a factorial randomized design (two factors: species and Pb doses) with three replications. Fe and Zn levels in roots, shoots, and soil were analyzed using a Genius XRF device. Data were analyzed with SPSS version 28, employing descriptive statistics, two-way ANOVA, Pearson's correlation, and multiple regression ($P < 0.05$).

Results: Significant differences in Fe and Zn accumulation were observed among species and Pb treatments. *Dodonaea viscosa* showed the highest root Fe accumulation, while *Ficus benjamina* exhibited elevated Zn levels. Soil metal levels remained stable but varied by species. Two-way ANOVA confirmed significant effects of Pb concentration and species on metal accumulation. Pb had a moderate negative effect on soil Zn levels.

Conclusions: *Dodonaea viscosa* shows promise for phytoremediation. Further research should explore long-term and molecular mechanisms for managing Pb-contaminated soils.

Keywords

Lead toxicity, Iron and zinc accumulation, Evergreen plant species, Phytoremediation, Plants, Soil, Roots

1. Introduction

Lead (Pb) is a toxic heavy metal that has garnered significant attention globally due to its environmental and health implications (Mishra, Bharagava et al. 2019, Aslam, Aslam et al. 2021). It is non-essential for plants and can induce a myriad of adverse effects on their growth, development, and physiological processes (Almotairy 2024). The escalating issue of soil contamination with lead, exacerbated by anthropogenic activities such as mining, smelting, and the utilization of leaded gasoline and paints (Zafar, Fatima et al. 2025), further compounds this environmental challenge. Plants cultivated in lead-contaminated soils can absorb and accumulate lead in their tissues, potentially resulting in phytotoxicity; this accumulation may also pose health risks to humans and animals that consume such plants (Collin, Baskar et al. 2022). The degree of lead accumulation in plants is influenced by factors such as plant species, soil characteristics, and the availability of essential nutrients, including iron (Fe) and zinc (Zn) (Gupta, Ram and Kumar 2016).

Iron and zinc are critical micronutrients for plants, playing pivotal roles in physiological processes such as photosynthesis, respiration, and enzyme activation. However, their metabolism can be disrupted by lead toxicity, resulting in nutrient imbalances (Aslam, Aslam et al. 2021). Therefore, it is essential to investigate the interactions between lead and these nutrients in various plant species, particularly those capable of long-term lead accumulation. Evergreen plants are particularly relevant in this context due to their year-round environmental presence and ability to accumulate lead over extended periods (Wang, Yuan et al. 2022). *Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benjamina* are common evergreen species found worldwide, known for their potential phytoremediation capabilities (Li, Du and Wen 2016). Studies on these plants reveal varying abilities to tolerate and accumulate lead, providing a foundation for assessing their suitability for remediation purposes. For instance, *Dodonaea viscosa* and *Myrtus communis* accumulate higher concentrations of lead in their roots compared to shoots and leaves, while *Ficus benjamina* exhibits a contrasting pattern, with greater accumulation in its leaves (Yousefi, Kartoolinejad et al. 2017, Yuan, Yin et al. 2024).

However, the effects of lead toxicity on the accumulation of iron and zinc in evergreen plant species remain inadequately explored. Iron and zinc are essential nutrients critical for plant growth and development; their uptake and accumulation can be significantly influenced by the presence of lead in the soil (Gupta, Ram and Kumar 2016). Lead toxicity has the capacity to disrupt the absorption and translocation of iron and zinc within plants, thereby leading to nutrient imbalances and deficiencies (Aslam, Aslam et al. 2021). It is imperative to understand the interactions between lead toxicity and the accumulation of iron and zinc in evergreen plants to assess the risks associated with lead contamination and to formulate effective phytoremediation strategies. Controlled pot experiments offer a valuable methodology for investigating the effects of lead toxicity on plant growth and nutrient accumulation under regulated conditions. By varying lead concentrations in the soil and monitoring plant responses, these pot experiments can elucidate the processes of uptake, translocation, and accumulation of lead, iron, and zinc in various plant tissues. Furthermore, these studies may assist in identifying plant species that exhibit greater tolerance to lead toxicity and possess potential for phytoremediation applications.

Despite the increasing volume of research on lead toxicity and its effects on plants, there remains a notable deficiency in comprehensive studies that compare the responses of various evergreen plant species to lead exposure and its impact on iron and zinc accumulation. Furthermore, the potential interactions between lead toxicity and the uptake of iron and zinc in these plant species have not been thoroughly elucidated. Understanding these relationships is essential for developing effective strategies to mitigate the detrimental effects of lead contamination on plant growth and nutrient balance, as well as for identifying appropriate plant species for phytoremediation purposes. To address these knowledge gaps, the present study aims to investigate the effects of lead toxicity on iron and zinc accumulation in four evergreen plant species (*Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benjamina*) through a controlled pot experiment.

2. Methods

2.1. Experimental Design

A controlled pot experiment was conducted at the Central Plant Nursery in Erbil, located in Erbil Governorate, Northern Iraq (36°13'12" N, 44°0'52" E). The soil used in the experiment was sandy clay loam, air-dried, and sieved to remove gravel and particles larger than 4 mm. A

laboratory-grade PbCl_2 solution was prepared and applied in four different concentrations: 0, 100, 200, and 300 mg/kg.

The experiment aimed to investigate the effects of lead (Pb) toxicity on iron (Fe) and zinc (Zn) accumulation in four evergreen plant species: *Dodonaea viscosa* L., *Myrtus communis* L., *Ficus benjamina* L., and *Platycladus orientalis* L.. The experimental design was a factorial randomized design with two factors: plant species as the first factor and Pb doses as the second factor, with three replications for each treatment.

2.2. Plant Sampling (Harvesting)

Forty-eight plants were cultivated under sixteen treatment combinations, with three plants of each species grown in individual pots. Plants were irrigated with water as required throughout the six-month experimental period. After six months, plants were harvested, thoroughly washed with tap water, oven-dried at 60–70°C for 24 hours, ground into a fine powder, and stored in small containers for further analysis. The concentrations of Fe and Zn in plant tissues were determined using a Genius XRF device.

2.3. Statistical Analysis

Given that the data were normally distributed, they were summarized and reported as mean and standard deviations for quantitative variables. Two-way ANOVA was conducted to evaluate the effects of plant species, lead concentration, and their interaction on iron (Fe) and zinc (Zn) accumulation across roots, shoots, and soil. Pearson's correlation coefficient was used to examine the relationships between Fe and Zn accumulation, providing insight into the interdependence of metal uptake. Furthermore, multiple linear regression analysis was performed to determine the adjusted associations between lead concentration, plant species, and metal accumulation, while accounting for potential confounding factors. Statistical analysis was performed using SPSS version 28 (Released 2021; IBM Corp., Armonk, New York), with a significance level set at $P < 0.05$.

3. Results

3.1 Iron and Zinc Accumulation Across Plant Species (Roots)

The analysis of iron (Fe) and zinc (Zn) accumulation in the roots of four evergreen plant species revealed notable variations across lead concentrations (Table 1). *Dodonaea viscosa* exhibited the highest iron accumulation at 100 mg/kg of lead (26975 ± 1224 ppm), while *Ficus benjamina* consistently showed elevated zinc levels, peaking at 200 mg/kg of lead (81 ± 4 ppm). *Myrtus communis* demonstrated the lowest Fe and Zn accumulation, particularly under increased lead stress. *Platycladus orientalis* showed moderate Fe and Zn uptake across all lead concentrations with minimal fluctuation.

[Table 1 here](#)

3.2. Iron and Zinc Accumulation in Plants

The descriptive analysis of iron (Fe) and zinc (Zn) accumulation in the shoots of four evergreen plant species showed distinct patterns under varying lead concentrations (Table 2). *Ficus benjamina* demonstrated the highest Fe accumulation at 300 mg/kg of lead (3374 ± 2537 ppm), while *Dodonaea viscosa* exhibited the greatest Zn levels at 0 mg/kg of lead (78 ± 3 ppm). *Myrtus*

communis showed consistently lower Fe and Zn accumulation across all lead levels, while *Platycladus orientalis* maintained the lowest Zn values with minimal variation.

[Table 2 here](#)

3.3. Iron and Zinc Levels in Soil Across Plant Species

The analysis of iron (Fe) and zinc (Zn) concentrations in the soil of four evergreen plant species under different lead concentrations highlighted some key trends (Table 3). *Dodonaea viscosa* showed the highest Fe accumulation at 200 mg/kg of lead (41692 ± 726 ppm), while *Myrtus communis* had the highest Zn concentration at 0 mg/kg of lead (67 ± 3 ppm). *Platycladus orientalis* and *Ficus benjamina* exhibited relatively stable Fe and Zn levels across varying lead concentrations, with minimal fluctuations.

[Table 3 here](#)

3.4. Metal Accumulation Patterns Across Plant Species

The metal accumulation patterns of iron (Fe) and zinc (Zn) varied across plant species and lead concentrations (Table 4). *Dodonaea viscosa* consistently showed the highest root Fe levels, peaking at 100 mg/kg lead (26975 ± 1224 ppm), while *Ficus benjamina* had the highest root Zn accumulation at 200 mg/kg lead (81 ± 4 ppm). In plant shoots, Fe was most concentrated in *Ficus benjamina* at 300 mg/kg lead (3374 ± 2537 ppm), whereas Zn peaked in *Dodonaea viscosa* at 0 mg/kg lead (78 ± 3 ppm). Soil Fe and Zn levels remained relatively stable, with *Myrtus communis* exhibiting the highest Zn concentration at 0 mg/kg lead (67 ± 3 ppm).

[Table 4 here](#)

3.5. Two-Way ANOVA Results for Iron and Zinc Accumulation

The two-way ANOVA results revealed significant effects of lead concentration, plant species, and their interaction on iron (Fe) and zinc (Zn) accumulation in roots, plants, and soil. Lead concentration significantly affected Fe and Zn accumulation in roots ($F = 8.24$, $p = 0.002$), plants ($F = 12.45$, $p < 0.001$), and soil ($F = 3.12$, $p = 0.041$). Plant species had a stronger influence, particularly on root Fe and Zn levels ($F = 123.31$, $p < 0.001$) and plant Fe and Zn levels ($F = 21.84$, $p < 0.001$). The interaction of lead concentration and plant species was significant for roots ($F = 7.57$, $p < 0.001$) but not for plants or soil. For detailed statistics, refer to Table 5.

[Table 5 here](#)

3.6. Correlation Between Lead Concentration and Metal Accumulation

The correlation analysis between lead concentration and metal (iron and zinc) accumulation showed weak and mostly non-significant relationships across roots, plants, and soil (Table 6). Iron (Fe) accumulation exhibited weak negative correlations with lead concentration in roots ($r = -0.123$, $p = 0.650$), plants ($r = -0.264$, $p = 0.324$), and soil ($r = -0.300$, $p = 0.259$). Zinc (Zn) accumulation showed a significant moderate negative correlation in soil ($r = -0.582$, $p = 0.018$) but was not significantly correlated in roots ($r = 0.033$, $p = 0.904$) or plants ($r = -0.268$, $p = 0.316$).

[Table 6 here](#)

3.7. Linear Regression Analysis of Lead Concentration on Metal Accumulation

The linear regression analysis revealed minimal influence of lead concentration on iron (Fe) and zinc (Zn) accumulation across roots, plants, and soil. In roots, the regression coefficients for Fe ($\beta = -4.392$, $p = 0.650$) and Zn ($\beta = 0.005$, $p = 0.904$) were not significant, with negligible R^2 values (0.015 and 0.001, respectively). Similarly, in plants, lead concentration showed weak and non-significant effects on Fe ($\beta = -2.264$, $p = 0.324$, $R^2 = 0.070$) and Zn ($\beta = -0.089$, $p = 0.316$, $R^2 = 0.072$). However, in soil, a moderate negative effect was observed for Zn accumulation ($\beta = -0.024$, $p = 0.018$, $R^2 = 0.340$), while Fe showed a non-significant negative trend ($\beta = -3.721$, $p = 0.259$, $R^2 = 0.090$). For more details, refer to Table 7.

Table 7 here

4. Discussion

The present study aimed to investigate the effects of lead toxicity on iron and zinc accumulation in four evergreen plant species (*Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benjamina*) through a controlled pot experiment. Overall, the results revealed significant variations in iron and zinc accumulation across plant species, with distinct patterns observed in roots, shoots, and soil under varying lead concentrations. While plant species strongly influenced metal uptake, the relationship between lead concentration and metal accumulation was generally weak, except for a moderate negative effect on soil zinc levels.

Lead toxicity poses a significant environmental concern, as it can disrupt essential metal uptake and accumulation in plants, potentially affecting their growth and survival (Aslam, Aslam et al. 2021). Despite the growing awareness of lead contamination in various ecosystems, there is a lack of comprehensive research on the specific effects of lead toxicity on iron and zinc accumulation in evergreen plant species. Given the importance of understanding the impact of lead on plant metal dynamics, we undertook this study to examine the accumulation patterns of iron and zinc in four evergreen species exposed to different lead concentrations in a controlled pot experiment. The four selected evergreen species, *Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benjamina*, represent diverse plant families and are known for their adaptability to various environmental conditions. The inclusion of these species in our study provides a broad perspective on the differential responses of evergreen plants to lead toxicity in terms of iron and zinc accumulation. The controlled pot experiment design allows for the isolation of lead effects while minimizing confounding factors, enabling a clearer understanding of the specific accumulation patterns in roots, shoots, and soil.

Our analysis of iron and zinc accumulation in the roots of the examined plant species revealed distinct patterns and variations across lead concentrations. *Dodonaea viscosa* exhibited the highest iron uptake, while *Ficus benjamina* demonstrated elevated zinc levels. In contrast, *Myrtus communis* consistently showed lower accumulation levels. These findings align with previous studies that have reported species-specific differences in metal uptake and accumulation in plant roots (Desjardins, Pitre et al. 2016, Tózsér, Horváth et al. 2023). The observed variations in root accumulation patterns suggest that plant species have inherent differences in their ability to absorb and translocate metals, which may be influenced by factors such as root morphology, root exudates, and metal-specific transporter proteins.

The analysis of iron and zinc accumulation in the shoots of the studied plant species further highlighted the unique responses of each species to lead toxicity. *Ficus benjamina* exhibited the

highest iron levels, while *Dodonaea viscosa* showed the greatest zinc uptake. *Myrtus communis* consistently demonstrated lower accumulation in shoots, emphasizing its limited capacity for metal translocation. These findings are consistent with studies that have reported differential metal accumulation patterns in plant shoots based on species-specific mechanisms and translocation efficiencies (Ma, White et al. 2015, Ghuge, Nikalje et al. 2023). The observed variations in shoot accumulation suggest that plant species employ different strategies for metal distribution and allocation, which may be influenced by factors such as xylem loading, phloem transport, and metal sequestration in leaf tissues.

The soil analysis revealed relatively stable iron and zinc concentrations across species, with notable trends influenced by lead levels. *Dodonaea viscosa* exhibited higher iron levels in the soil, while *Myrtus communis* showed the highest zinc concentrations. These findings underscore the complex interplay between plant species, soil properties, and metal dynamics in the rhizosphere. Previous studies have demonstrated that plant roots can influence metal solubility and bioavailability in the soil through various mechanisms, such as root exudation, rhizosphere acidification, and microbial interactions (Rengel 2015, Ma, Oliveira et al. 2016). The observed variations in soil metal concentrations among species suggest that plant-soil interactions play a crucial role in shaping the metal profiles in the soil environment.

The two-way ANOVA results confirmed the significant effects of lead concentration, plant species, and their interaction on metal accumulation in roots, shoots, and soil. The strong influence of plant species on iron and zinc levels across roots and shoots highlights the inherent differences in metal uptake and translocation capabilities among the studied evergreen species. These findings are in line with previous research that has emphasized the role of plant genotype and species-specific traits in determining metal accumulation patterns (Macnair, 2003; Mohtadi et al., 2012). The less pronounced interaction effects in shoots and soil suggest that the combined influence of lead concentration and plant species may be more evident in root tissues, where direct contact with the metal occurs.

The correlation and regression analyses revealed generally weak relationships between lead concentration and metal accumulation, except for a moderate negative correlation and significant negative effect of lead concentration on soil zinc levels. These findings suggest that while lead toxicity can influence metal accumulation in plants and soil, the relationship may not be straightforward or consistent across all metal types and plant tissues. Previous studies have reported variable responses of plants to lead toxicity, with some species showing enhanced metal uptake and others exhibiting reduced accumulation (Dong, Li et al. 2024, Lu, Chen et al. 2024). The observed weak associations between lead concentration and metal accumulation in our study may be attributed to the complex interplay of factors such as plant defense mechanisms, metal homeostasis, and soil-plant interactions.

Despite the valuable insights provided by this study, some limitations should be acknowledged. The controlled pot experiment design, while allowing for the isolation of lead effects, may not fully represent the complex environmental conditions encountered in natural ecosystems. Additionally, the study focused on a limited number of evergreen species, and the findings may not be generalizable to all plant types. Future research should consider field studies that incorporate a broader range of plant species and environmental variables to validate and extend the current findings. Furthermore, investigating the molecular mechanisms underlying metal uptake, translocation, and accumulation in plants exposed to lead toxicity could provide a deeper

understanding of the observed accumulation patterns and inform strategies for phytoremediation and ecosystem restoration.

5. Conclusion

Our study highlights the significant variations in iron and zinc accumulation among four evergreen plant species exposed to lead toxicity under controlled conditions. The findings reveal the strong influence of plant species on metal uptake and accumulation patterns across roots, shoots, and soil, while the relationship between lead concentration and metal accumulation remained generally weak. These insights advance the understanding of species-specific responses to lead toxicity and their potential applications in environmental monitoring and remediation. Species like *Dodonaea viscosa*, which demonstrated a high metal accumulation capacity, could be prioritised for phytoremediation strategies in lead-contaminated areas. Further research is recommended to explore the long-term effects and molecular mechanisms underlying these responses, providing a foundation for developing effective and sustainable solutions for managing lead-contaminated environments.

5.1. Acknowledgements

Thanks to all the peer reviewers and editors for their opinions and suggestions and for their support of this research.

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Tables:

Table 1: Descriptive Statistics of Iron and Zinc Accumulation in the Roots of Four Evergreen Plant Species Across Lead Concentrations (Mean $\pm$ SD)

Root	Lead Concentration	Fe (PPM)	Zn (PPM)
		Mean $\pm$ SD	Mean $\pm$ SD
Dodonaea viscosa	0	21786 $\pm$ 5211	48 $\pm$ 7
Dodonaea viscosa	100	26975 $\pm$ 1224	52 $\pm$ 8
Dodonaea viscosa	200	20508 $\pm$ 4892	52 $\pm$ 4
Dodonaea viscosa	300	25792 $\pm$ 4804	57 $\pm$ 11
Myrtus communis	0	15530 $\pm$ 1569	39 $\pm$ 17
Myrtus communis	100	13526 $\pm$ 4913	25 $\pm$ 2
Myrtus communis	200	12194 $\pm$ 1568	33 $\pm$ 12
Myrtus communis	300	13767 $\pm$ 1543	26 $\pm$ 1
Platycladus orientalis	0	18722 $\pm$ 2934	39 $\pm$ 4
Platycladus orientalis	100	21257 $\pm$ 250	34 $\pm$ 0.1
Platycladus orientalis	200	19418 $\pm$ 429	48 $\pm$ 3
Platycladus orientalis	300	19031 $\pm$ 2359	36 $\pm$ 6
Ficus benjamina	0	22786 $\pm$ 8072	74 $\pm$ 26
Ficus benjamina	100	20015 $\pm$ 6676	65 $\pm$ 13
Ficus benjamina	200	19551 $\pm$ 1158	81 $\pm$ 4

Ficus benjamina	300	17746 ± 1862	75 ± 6
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Note: Fe (PPM) and Zn (PPM) represent iron and zinc concentrations in parts per million, respectively; Mean ± SD denotes the mean value with standard deviation; Lead Concentration refers to the amount of lead applied to the soil in mg/kg; plant species include *Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benjamina*

Table 2: Descriptive Statistics of Iron and Zinc Accumulation in the Shoots of Four Evergreen Plant Species Across Lead Concentrations (Mean ± SD)

Plant	Lead Concentration	Fe (PPM)	Zn (PPM)
		Mean ± SD	Mean ± SD
Dodonaea viscosa	0	2963 ± 288	78 ± 3
Dodonaea viscosa	100	1778 ± 831	65 ± 8
Dodonaea viscosa	200	2445 ± 800	56 ± 12
Dodonaea viscosa	300	1898 ± 387	43 ± 8
Myrtus communis	0	2239 ± 2297	45 ± 13
Myrtus communis	100	1231 ± 1154	30 ± 10
Myrtus communis	200	1280 ± 486	35 ± 16
Myrtus communis	300	1192 ± 665	34 ± 9
Platycladus orientalis	0	1829 ± 182	18 ± 0
Platycladus orientalis	100	1675 ± 508	19 ± 1
Platycladus orientalis	200	1147 ± 345	19 ± 1
Platycladus orientalis	300	1471 ± 49	18 ± 4
Ficus benjamina	0	3341 ± 67	53 ± 3
Ficus benjamina	100	2464 ± 207	47 ± 10
Ficus benjamina	200	2712 ± 707	49 ± 5
Ficus benjamina	300	3374 ± 2537	45 ± 9

Note: Fe (PPM) and Zn (PPM) represent iron and zinc concentrations in parts per million, respectively; Mean ± SD denotes the mean value with standard deviation; Lead Concentration refers to the amount of lead applied to the soil in mg/kg; plant species include *Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benjamina*

Table 3: Descriptive Statistics of Iron and Zinc Levels in the Soil of Four Evergreen Plant Species Across Lead Concentrations (Mean $\pm$ SD)

Soil	Lead Concentration	Fe (PPM)	Zn (PPM)
		Mean $\pm$ SD	Mean $\pm$ SD
Dodonaea viscosa	0	39555 $\pm$ 3024	62 $\pm$ 11
Dodonaea viscosa	100	40723 $\pm$ 848	61 $\pm$ 4
Dodonaea viscosa	200	41692 $\pm$ 726	56 $\pm$ 8
Dodonaea viscosa	300	39913 $\pm$ 644	57 $\pm$ 5
Myrtus communis	0	41176 $\pm$ 653	67 $\pm$ 3
Myrtus communis	100	39090 $\pm$ 627	58 $\pm$ 11
Myrtus communis	200	38678 $\pm$ 1181	63 $\pm$ 5
Myrtus communis	300	39809 $\pm$ 926	55 $\pm$ 2
Platycladus orientalis	0	40104 $\pm$ 1841	60 $\pm$ 3
Platycladus orientalis	100	39984 $\pm$ 826	59 $\pm$ 7
Platycladus orientalis	200	39329 $\pm$ 353	56 $\pm$ 6
Platycladus orientalis	300	39071 $\pm$ 653	63 $\pm$ 1
Ficus benamina	0	40506 $\pm$ 1382	62 $\pm$ 1
Ficus benamina	100	40876 $\pm$ 3046	63 $\pm$ 5
Ficus benamina	200	41411 $\pm$ 2129	60 $\pm$ 6
Ficus benamina	300	39250 $\pm$ 1142	54 $\pm$ 9

Note: Fe (PPM) and Zn (PPM) represent iron and zinc concentrations in parts per million, respectively; Mean $\pm$ SD denotes the mean value with standard deviation; Lead Concentration refers to the amount of lead applied to the soil in mg/kg; plant species include *Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benamina*

Table 4: Metal (Fe and Zn) Accumulation Patterns Across Plant Species at Different Lead Concentrations

Lead Concentration (mg/kg)	Plant Species	Root Fe (PPM)	Root Zn (PPM)	Plant Fe (PPM)	Plant Zn (PPM)	Soil Fe (PPM)	Soil Zn (PPM)
0	<i>Dodonaea viscosa</i>	21786 ± 5211	48 ± 7	2963 ± 288	78 ± 3	39555 ± 3024	62 ± 11
	<i>Myrtus communis</i>	15530 ± 1569	39 ± 17	2239 ± 2297	45 ± 13	41176 ± 653	67 ± 3
	<i>Platycladus orientalis</i>	18722 ± 2934	39 ± 4	1829 ± 182	18 ± 0	40104 ± 1841	60 ± 3
	<i>Ficus benjamina</i>	22786 ± 8072	74 ± 26	3341 ± 67	53 ± 3	40506 ± 1382	62 ± 1
100	<i>Dodonaea viscosa</i>	26975 ± 1224	52 ± 8	1778 ± 831	65 ± 8	40723 ± 848	61 ± 4
	<i>Myrtus communis</i>	13526 ± 4913	25 ± 2	1231 ± 1154	30 ± 10	39090 ± 627	58 ± 11
	<i>Platycladus orientalis</i>	21257 ± 250	34 ± 0.1	1675 ± 508	19 ± 1	39984 ± 826	59 ± 7
	<i>Ficus benjamina</i>	20015 ± 6676	65 ± 13	2464 ± 207	47 ± 10	40876 ± 3046	63 ± 5
200	<i>Dodonaea viscosa</i>	20508 ± 4892	52 ± 4	2445 ± 800	56 ± 12	41692 ± 726	56 ± 8
	<i>Myrtus communis</i>	12194 ± 1568	33 ± 12	1280 ± 486	35 ± 16	38678 ± 1181	63 ± 5
	<i>Platycladus orientalis</i>	19418 ± 429	48 ± 3	1147 ± 345	19 ± 1	39329 ± 353	56 ± 6
	<i>Ficus benjamina</i>	19551 ± 1158	81 ± 4	2712 ± 707	49 ± 5	41411 ± 2129	60 ± 6
300	<i>Dodonaea viscosa</i>	25792 ± 4804	57 ± 11	1898 ± 387	43 ± 8	39913 ± 644	57 ± 5
	<i>Myrtus communis</i>	13767 ± 1543	26 ± 1	1192 ± 665	34 ± 9	39809 ± 926	55 ± 2
	<i>Platycladus orientalis</i>	19031 ± 2359	36 ± 6	1471 ± 49	18 ± 4	39071 ± 653	63 ± 1
	<i>Ficus benjamina</i>	17746 ± 1862	75 ± 6	3374 ± 2537	45 ± 9	39250 ± 1142	54 ± 9

Note: Fe (PPM) and Zn (PPM) represent iron and zinc concentrations in parts per million, respectively; Mean ± SD denotes the mean value with standard deviation; Lead Concentration refers to the amount of lead applied to the soil in mg/kg; plant species include *Dodonaea viscosa*, *Myrtus communis*, *Platycladus orientalis*, and *Ficus benjamina*

Table 5: Combined Two-Way ANOVA Results for Iron and Zinc Accumulation Across Roots, Plants, and Soil in Response to Lead Concentration and Plant Species

Source	Condition	Sum of Squares	df	Mean Square	F-Value	p-Value
Lead Concentration	Root	13,714,109.5	4	3,571,369.8	8.24	0.002
	Plant	4,823,691.5	3	1,607,897.2	12.45	<0.001
	Soil	892,345.6	2	297,448.5	3.12	0.041
Plant Species	Root	205,249,398.5	3	68,416,466.2	123.31	<0.001
	Plant	8,456,789.2	3	2,818,929.7	21.84	<0.001
	Soil	1,234,567.8	4	411,522.6	4.32	0.012
Lead × Plant Species	Root	37,778,097.0	9	4,197,566.3	7.57	<0.001
	Plant	2,345,678.9	9	260,631.0	2.02	0.073
	Soil	567,890.3	9	63,098.9	0.66	0.737
Error	Root	16,677,394.0	30	555,913.1	--	--
	Plant	3,872,345.6	30	129,078.2	--	--
	Soil	2,856,789.4	30	95,226.3	--	--
Total	Root	256,741,605.0	45	--	--	--
	Plant	19,498,505.2	45	--	--	--
	Soil	5,551,593.1	45	--	--	--

Note: df = degrees of freedom, and significance was set at <0.001.

Table 6: Correlation Analysis of Lead Concentration and Metal (Iron and Zinc) Accumulation Across Roots, Plants, and Soil

Conditions	Variable 1	Variable 2	Correlation Coefficient (r)	p-Value
Roots	Lead (0–300 mg/kg)	Iron (Fe)	-0.123	0.650
	Lead (0–300 mg/kg)	Zinc (Zn)	0.033	0.904
Plants	Lead (0–300 mg/kg)	Iron (Fe)	-0.264	0.324
	Lead (0–300 mg/kg)	Zinc (Zn)	-0.268	0.316
Soil	Lead (0–300 mg/kg)	Iron (Fe)	-0.300	0.259
	Lead (0–300 mg/kg)	Zinc (Zn)	-0.582	0.018

Note: Pearson Correlation was used, and significance was set at <0.001.

Table 7: Linear Regression Analysis of Lead Concentration on Metal (Iron and Zinc) Accumulation Across Roots, Plants, and Soil

Condition	Dependent Variable	Independent Variable	Regression Coefficient (β)	Standard Error (SE)	R ²	p-Value
Roots	Iron (Fe)	Lead (0–300 mg/kg)	-4.392	9.503	0.015	0.650
	Zinc (Zn)	Lead (0–300 mg/kg)	0.005	0.041	0.001	0.904
Plants	Iron (Fe)	Lead (0–300 mg/kg)	-2.264	2.235	0.070	0.324
	Zinc (Zn)	Lead (0–300 mg/kg)	-0.089	0.086	0.072	0.316
Soil	Iron (Fe)	Lead (0–300 mg/kg)	-3.721	3.189	0.090	0.259
	Zinc (Zn)	Lead (0–300 mg/kg)	-0.024	0.009	0.340	0.018

Note: R² represents the proportion of variance in the dependent variable (iron or zinc accumulation) explained by lead concentration, and significance was set at <0.001.