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Mitigation Potential of Vermicompost on Morphophysiological and Mineral Responses of Rocket (Eruca sativa L.) Under High Lead (Pb) Stress

Sultan Dere Uğur[1,a,*]

[1]*Siirt University, Faculty of Agriculture, Department of Horticulture, 56000, Siirt, Turkey*
[*]*Corresponding author*

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ABSTRACT

Heavy metal pollution, one of the abiotic stress factors, negatively affects agricultural production. Lead (Pb) is one of the most important heavy metals affecting agricultural production. It is important to determine the effect of lead on rocket (Eruca sativa L.), which has a high cultivation potential in agricultural production, and to investigate the effect of vermicompost application in reducing the negative effect of lead stress. This study was carried out in a climate chamber using solid media culture. Rocket (Eruca sativa L.) cv. Bengi was used as plant material. The experiment included four treatments: control (no treatment), vermicompost treatment, 1500 mg kg^{-1} lead stress, and 1500 mg kg^{-1} lead stress + vermicompost treatment. The study was terminated after the effects of lead stress were observed. Pb stress increased plant height from 27.42 cm (control) to 25.92 cm, while a greater reduction was observed under Pb stress + vermicompost (24.95 cm). Plant fresh weight declined from 55.23 g in the control to 26.17 g under Pb stress + vermicompost. Plant dry weight decreased from 3.40 g (control) to 1.61 g in the combined treatment. SPAD values increased from 42.10 (control) to 50.52 with vermicompost, while Pb stress resulted in a moderate increase (44.60). Ion leakage increased from 9.76% (control) to 15.33% under Pb stress and reached a maximum of 23.57% in the Pb stress + vermicompost treatment. Leaf Pb content increased from 1.31 ppm (control) to 4.13 ppm under Pb stress, but decreased when vermicompost was applied together with Pb. The results demonstrate that Pb stress disrupts growth, membrane integrity, and nutrient homeostasis in rocket plants. Lead stress severely impairs growth and physiological performance of rocket plants. Vermicompost application alone exerted positive effects on certain physiological traits; however, under Pb stress conditions, it was insufficient to mitigate Pb toxicity and, in some parameters, intensified stress responses.

[a] *sultan.dere@siirt.edu.tr* *https://orcid.org/0000-0001-5928-1060*

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Introduction

Rocket (*Eruca sativa* L.) is a cool-season leafy vegetable belonging to the Brassicaceae family and is widely accepted to have originated in the Mediterranean region (Özkan and Ünlü, 2024). The optimal temperature range for rocket growth is 15–20°C, while temperatures below 10°C adversely affect germination and early development. Under favorable environmental conditions, particularly during summer, the period from sowing to harvest is shortened; however, exposure to low temperatures may enhance the accumulation of aromatic compounds at the expense of overall quality. Rocket performs best during spring and autumn when adequate soil and atmospheric humidity are maintained and extreme temperatures are avoided (Hall et al. 2012). Owing to its high nutritional value and reported aphrodisiac properties, rocket consumption has increased substantially, and it is increasingly incorporated into various dietary and therapeutic applications (Al-Rawe et al. 2023).

In recent decades, heavy metal contamination has emerged as a major global environmental and agricultural concern due to its persistence, toxicity, and capacity for accumulation in soils and biological systems. Anthropogenic activities—including industrial emissions, mining operations, fossil fuel combustion, wastewater discharge, and intensive agrochemical use—have been identified as the dominant contributors to heavy metal pollution in terrestrial ecosystems, far exceeding natural geogenic sources. Heavy metals such as cadmium (Cd), lead (Pb), mercury (Hg), arsenic (As), chromium (Cr), nickel (Ni), copper (Cu), and zinc (Zn) can persist in soils for extended periods and enter the food chain, thereby threatening soil health, crop productivity, and human well-being (Angon et al. 2024).

Among heavy metals, lead (Pb) is considered one of the most hazardous environmental contaminants due to its high toxicity to ecosystems and living organisms. The global production and environmental dissemination of Pb continue to increase in response to growing demand from battery manufacturing, the automotive industry, and other industrial applications (Raj and Das, 2023; Sweta and Singh, 2022). As a non-essential element, Pb persists in soil, water, and air, leading to long-term ecological risks and a decline in soil fertility (Ahmad et al. 2021; Nieder and Benbi, 2024). Pb-contaminated soils exhibit reduced microbial diversity and enzymatic activity, which impairs nutrient cycling and ultimately restricts plant growth and agricultural productivity (Lugun et al. 2022; Botté et al. 2022). Furthermore, the tendency of Pb to bioaccumulate and biomagnify along the food chain intensifies its ecological and human health impacts (Meena et al. 2019; Collin et al. 2022).

Elevated Pb concentrations negatively affect seed germination, plant growth, and metabolic processes, resulting in substantial yield and quality losses. Pb toxicity induces oxidative stress by triggering excessive production of reactive oxygen species (ROS), including hydroxyl radicals, superoxide anions, and hydrogen peroxide, which cause cellular and biochemical damage (Osman and Fadhlallah, 2023; Berni et al. 2019). Although plants activate enzymatic and non-enzymatic antioxidant systems to counteract Pb-induced oxidative damage, these defense mechanisms are often insufficient under prolonged or high Pb exposure. Consequently, the development of environmentally friendly and sustainable strategies to mitigate Pb stress in agricultural systems has become imperative (Collin et al. 2022; Shan et al. 2023).

The rapid growth of the global population, coupled with the continuous reduction of arable land, poses significant challenges to food production (Lal, 2009). To meet increasing food demands, the intensive use of chemical fertilizers has become widespread; however, this practice has resulted in unsustainable agricultural systems, declining soil fertility, environmental pollution, and a reduction in beneficial soil microorganisms (Cakmakçı and Erdoğan, 2008). To address these issues, the use of organic fertilizers, such as vermicompost, is gaining traction (Dere, 2020).

Vermicompost is a valuable organic amendment characterized by its porous structure, which enhances soil water-holding capacity, aeration, and drainage.

It is rich in essential nutrients such as nitrogen, phosphorus, and potassium, as well as beneficial microorganisms (Garg et al. 2006; Tavuç and Özçelik, 2015). These microorganisms secrete enzymes and plant growth regulators, including auxins, cytokinins, and gibberellic acid, into the rhizosphere, thereby promoting nutrient uptake, suppressing soil-borne pathogens, and improving plant growth, yield, and quality (Arancon et al. 2003; Sharma and Banik, 2014). Moreover, vermicompost has been reported to enhance plant tolerance to abiotic stresses such as drought by influencing protein and enzyme synthesis, leading to improved growth performance under stress conditions (Muscolo et al. 2007; Hosseinzadeh et al. 2016; Cobanoglu, 2022).

In this context, increasing Pb pollution resulting from industrial and agricultural activities represents a serious threat to sustainable crop production. Therefore, identifying effective strategies to mitigate Pb stress in plants is of critical importance. The present study aims to evaluate the morphological and physiological responses of the Bengi rocket cultivar to lead contamination at a concentration of 1500 mg kg^{-1} and vermicompost application. The findings are expected to contribute to the efficient utilization of diminishing agricultural lands, enhancement of soil organic matter content, and maintenance of agricultural productivity under increasing environmental stress conditions.

Materials and Methods

The experiment was conducted in the climate room of the Department of Field Crops, Siirt University. The climate room of the Department of Field Crops has a temperature range of -20°C and +40°C, light intensity of 400 µmol $m^{-2}s^{-1}$, photoperiodicity (day/night) and humidity conditions can be adjusted as desired. For rocket (*Eruca sativa* L.) cultivation, an average temperature of 15-20±2 ºC, 70% humidity and 16 hours of light and 8 hours of darkness were set. Bengi rocket variety was used as plant material in our study. The seeds used in the experiment were obtained from Malkoç Agricultural Company. This study utilized liquid vermicompost fertilizer produced by Yeşil Vadisi Agriculture.

Our study was organized according to the randomized plots experimental design with 3 replications and 10 plants in each replicate. There were 12 plots in total, 10 plants in each plot and 120 plants in the experiment.

Applications

1. Control (No treatment; peat: perlite mixture at a ratio of 3:1, v:v)
2. Vermicompost application (vermicompost: peat: perlite mixture at a ratio of 1:2:1, v:v:v)
3. 1500 mg kg-1 heavy metal Pb stress treatment (peat: perlite mixture at 3:1, v:v ratio)
4. 1500 mg kg-1 heavy metal Pb stress application (vermicompost: peat: perlite mixture at a ratio of 1:2:1, v:v:v). The heavy metal concentration used in our study was determined according to the study by Balcı (2019). Vermicompost rate was based on the dose recommended by the commercial company. The content of vermicompost purchased from a commercial company is given in Table 1.

Rocket seeds were sterilized using 5% sodium hypochlorite. The sterilized seeds were washed 3 times with distilled water to remove hypochlorite. Then, rocket seeds were sown at a depth of 0.5 cm by volume in vials containing peat: perlite mixture mixed at a ratio of 3:1. After the seeds were sown and necessary controls were made, they were watered with tap water. After germination of the seeds, the standard nutrient solution used during the study was diluted and irrigated to the plants at the seedling stage when necessary. This standard nutrient solution contained [(M: 3.0 $x10^{-3}$), $Ca(NO_3)_2$; $0.9x10^{-3}$, K_2SO_4; $1.0x10^{-3}$, $MgSO_4$; $0.2x10^{-3}$, KH_2PO_4; $1.0x10^{-5}$, H_3BO_3; $1.0x10^{-6}$, $MnSO_4$; $1.0x10^{-7}$, $CuSO_4$; $1.0x10^{-8}$, $ZnSO_4$; $1.0x10^{-4}$, $(NH_4)_6Mo_7O_{24}$; $1.0x10^{-6}$ and Fe-EDTA (10^{-4} M)] (Hoagland and Arnon, 1950). According to the applications, 2 L pots were prepared volumetrically.

Table 1. Vermicompost content used in the experiment

Analysis name	Vermicompost	Analysis name	Vermicompost
Dry matter (%)	44.32	Ammonium nitrogen (%)	0.130
Moisture (%)	55.68	Nitrate nitrogen (%)	0.340
Organic matter (%)	38.70	Water soluble potassium (%)	0.610
pH (1 10^{-1})	7.12	Water soluble phosphorus (%)	0.390
EC (1 10^{-1}, dSm^{-1})	1.37	Water soluble CaO (%)	3.970
Total nitrogen (%)	3.34	Water soluble MgO (%)	6.590
Total phosphorus (%)	1.74	Water soluble Cl (%)	0.068
Total potassium (%)	0.99	Water soluble SO_4 (%)	1.330
Total CaO (%)	0.15	Water soluble sodium (%)	0.022
Total MgO (%)	0.37	Total boron (ppm)	20.000

Plants were transplanted into these pots when the first true leaves appeared (seedling stage with two true leaves). There were 2 plants in each pot. Plants were irrigated with Hoagland's nutrient solution given above until they reached the treatment stage. Sample pots were kept to determine the amount and time of watering for each treatment. For each application, the pots were placed in a container and watered from the top to reach the saturation point, and the amount of irrigation was calculated by taking into account the amount of water drained. Drainage levels were determined daily and this rate was set at approximately 30% during the experiment. Then the necessary preparations were made for the applications.

Heavy metal Pb stress application was 1500 mg kg^{-1} (ppm). Heavy metal lead was divided according to the irrigation time in the heavy metal treated plots. Heavy metal application was started after the plants reached the two true-leafed stage, and heavy metal stress was started to be applied when the plants were shifted and taken into pots and the shock of shifting was over. Lead heavy metal added to the nutrient solution was given to the plants with irrigation based on the ratio of "drained solution/applied solution" (Schröder and Lieth, 2002). The pH and EC of the drained water were measured at the time of irrigation. The pH of the nutrient solution was kept in the range of 6.0-6.5 and EC in the range of 1.5-2.5 dS m^{-1}.

The experiment was terminated when the plants showed symptoms of heavy metal Pb stress. At the end of the experiment, the following analyzes and observations were made. Leaf samples were taken from the plants for enzyme analysis. Leaf samples were taken and placed in dry ice and then stored at -20°C.

Examined characteristics

0-5 Scale evaluation in green parts

In order to express the visible symptoms of Pb stress-induced damage in plants, a 0-5 scale was used. In heavy metal lead stress, a score from 0 to 5 was given according to the following symptoms (Daşgan et al. 2010; Kuşvuran et al. 2011).

1. Not affected at all (Control plants)
2. Slowdown in growth
3. Yellowing of the leaf blade on the lower leaves
4. Yellowing of the upper leaves between the veins (around 30% of the leaves)
5. Equivalent change in 80% or more of the leaves and onset of drying of the leaves
6. Complete yellowing of the plant and drying or death of the lower leaves

Determination of plant height and diameter (cm and mm)

At the end of the experiment, the area from the root collar to the tip of the plant was measured in cm (± 0.5) with a meter. Plant stem diameter was determined in mm (± 0.1) using a mechanical caliper.

Determination of leaf number (number $plant^{-1}$)

At the end of the experiment, all leaves on the plant were counted and the number of leaves was determined as number $plant^{-1}$.

Measurement with SPAD meter for chlorophyll

In order to determine the shade of green due to chlorophyll content in rocket plants grown under heavy metal Pb stress and control conditions, readings were taken with a Minolta SPAD meter.

Plant fresh and dry weights (g)

All plant samples were weighed and recorded. The weighed plant samples were dried at 70oC for approximately 48-72 hours and the dry weight was weighed after reaching constant weight.

Leaf relative water content (RWC)

Leaves of the same size and stage were used for the determination of leaf relative water content (RWC). Firstly, 4 leaf discs of 1 cm were weighed and recorded and these discs were kept in pure water for 4 hours and then weighed to determine the turgor weight. After the turgor weight was determined, the samples were dried in an oven at 70°C and the dry weight was determined (Illouz-Eliaz et al. 2025).

Relative water content was calculated according to the formula below.

$$RWC\ (\%) = [(W - DW)/(TW - DW)]\ x\ 100 \qquad (1)$$

RWC: Relative water content (%) W: Sample fresh weight (g), DW: Sample dry weight (g), TW: Sample turgid weight (g)

Ion leakage (%)

For each treatment, 1 cm diameter disks were taken from leaf samples of the same age and size. The disc samples were washed in distilled water and dried with blotting paper, then 3 discs were placed in tubes containing 20 ml of distilled water. The tubes containing the disc samples were shaken on a shaker for 4 hours and EC1 was measured with a conductivity meter (EC meter).

After the EC1 measurement, the tubes were kept in an autoclave at 121°C for 10 minutes, then the tubes were removed from the autoclave and allowed to cool at room temperature. EC2 was measured with a conductivity meter (EC meter) from the samples cooled at room temperature.

Ion leakage was calculated using the following equation (Gülen et al. 2007):

$$Ion\ leakage\ (\%) = (EC1/EC2)\ x\ 100 \qquad (2)$$

Using the following formula calculated as % compared to the control.

Tissue elemental analysis

In order to compare the nutritional status of the rocket (*Eruca sativa* L.) plants grown under different treatments, 50th day after transplanting, leaf analysis for P, K, Ca, Mg, Fe, Mn, Zn, and Cu were conducted. Samples of 20 leaves from each replication of all trails were collected. Rocket leaves were dried at 65°C for 48 h. After drying, samples were grinded using a mill with a 20 mesh sieve. The leaf powder was turned to ash at 550 °C for about 6 h and the ash was dissolved in 3.3% HCI. The concentrations of K, Ca, Mg, Fe, Mn, Zn, Cu in leaves were determined by atomic absorption spectrometry (Jones, 2001). Nitrogen concentration was determinated by Kjeldahl method (Kacar, 1995).

Statistical Analyses

Data were analyzed by analysis of variance (ANOVA) using JMP® version 7.0 statistical software (SAS Institute Inc. Cary, NC, USA). When the F-test was significant, mean comparisons were performed using the least significant difference (LSD) test. The LSD test was selected for its suitability in balanced experimental designs with a limited number of treatments and its sensitivity in detecting differences among treatment means at $p \leq 0.05$.

Results

The study investigated the effects of various treatments—control, vermicompost, 1500 ppm lead (Pb) stress, and 1500 ppm Pb stress combined with vermicompost—on several parameters such as 0-5 scale value, plant height, plant diameter, number of leaves, plant fresh weight, plant dry weight, root wet weight and root dry weight of the Bengi rocket variety (Table 2).

0–5 Scale Values

In terms of the 0–5 scale values (leaf damage), the statistical difference between the treatments was not found to be significant. In the evaluation conducted on a 0–5 scale, the lowest value was recorded in the control group (1.00), followed by the vermicompost treatment (1.50) and the 1500 ppm Pb stress treatment (1.83). The highest value was observed in the combined application of 1500 ppm Pb + vermicompost (2.50). These results indicate that Pb stress increased the severity of effects reflected on the scale, and that vermicompost application did not mitigate this effect.

Plant Height

No statistically significant differences were detected among the treatments with respect to plant height ($p > 0.05$). In terms of plant height, the highest value was obtained under the 1500 ppm Pb treatment (27.42 cm). The control (25.92 cm) and vermicompost (25.10 cm) treatments exhibited similar values. The lowest plant height was recorded in the Pb + vermicompost treatment, with a value of 24.95 cm.

Stem Diameter

When stem diameter results were examined, the highest value was measured in the 1500 ppm Pb treatment (3.77 mm). The control (3.51 mm) and Pb + vermicompost (3.50 mm) treatments exhibited similar values, whereas the vermicompost treatment resulted in the lowest stem diameter (3.31 mm). While some research highlights positive effects of vermicompost on plant growth, others report mixed or negative outcomes, suggesting that its impact can vary depending on the context (Blouin et al. 2019; Lazcano and Dominguez, 2010).

Number of Leaves

No statistically significant differences were detected among the treatments with respect to number of leaves ($p > 0.05$). In terms of the number of leaves per plant, the highest value was recorded in the control group (15.00 leaves plant^{-1}). A reduction in leaf number was observed in the vermicompost (12.83 leaves plant^{-1}), 1500 ppm Pb (12.17 leaves plant^{-1}), and Pb + vermicompost (12.00 leaves plant^{-1}) treatments (Table 2). The lowest leaf number was obtained in the Pb + vermicompost combination. This suggests that vermicompost did not alleviate the adverse effects of Pb stress on leaf number. Previous studies have similarly reported that Pb stress reduces leaf number in plants, aligning with our findings (Baktemur, 2022; Balcı, 2019; Kara et al. 2022; Opeolu et al. 2010).

Plant Fresh Weight

In terms of plant fresh weight, the highest value was recorded in the control group (55.23 g). Plant fresh weight decreased to 40.51 g under the vermicompost treatment, while a value of 48.73 g was determined under 1500 ppm Pb stress. The lowest plant fresh weight was measured in the 1500 ppm Pb + vermicompost combination (26.17 g). This supports earlier research indicating that Pb stress decreases biomass and that vermicompost can sometimes have mixed or negative effects on plant weight (Balcı, 2019; Lazcano and Dominguez, 2010). Although vermicompost can enhance growth by improving nutrient availability and hormone production (Azarmi et al. 2008; García Martinez et al. 2002), its effectiveness can be context-dependent.

Plant Dry Weight

Plant dry weight showed statistically significant differences among treatments. The control treatment produced the highest value (3.40 g), whereas plant dry weight decreased under vermicompost (2.58 g) and 1500 ppm Pb stress (2.72 g) applications. The lowest plant dry weight was recorded in the Pb + vermicompost treatment (1.61 g) (Table 2). While some studies have reported increased shoot dry weight with vermicompost (Paul and Metzger, 2005; Akhzari et al. 2016), others found no significant effect (Bachman and Metzger, 2007). Vermicompost has been noted to increase shoot dry weight in specific conditions, such as salt stress (Kıran, 2019), but its impact can vary.

Root Fresh Weight

No statistically significant differences were detected among the treatments with respect to root fresh weight ($p>0.05$). In terms of root fresh weight, the highest value was recorded in the control group (4.66 g). Root fresh weight was lower in the vermicompost (2.74 g), 1500 ppm Pb stress (2.58 g), and 1500 ppm Pb + vermicompost (2.64 g) treatments compared with the control. The reductions in root fresh weight observed in Pb-containing treatments were at similar levels. Previous research supports these findings, indicating that Pb stress can decrease root weight (Balcı, 2019), while moderate Pb concentrations might enhance root growth (Kara et al. 2022).

Table 2. Effects of control, vermicompost, Pb stress and Pb stress+vermicompost treatments on some plant growth parameters in rocket plants

Treatment	0-5 SV	PH (cm)	SD (mm)	NL	PFW (g)	PDW(g)	RFW (g)	RDW(g)
Control	1.00± 0.00	25.92± 0.35	3.51± 0.41	15.00± 1.41	55.23± 11.92^{a}	3.40± 0.94^{a}	4.66± 0.21	0.49± 0.01^{a}
Vermicompost	1.50± 0.35	25.10± 7.07	3.31± 0.01	12.83± 3.54	40.51± 7.71^{b}	2.58± 0.58^{b}	2.74± 0.24	0.27± 0.01^{b}
1500 ppm Pb stress	1.83± 0.71	27.42± 0.78	3.77± 0.90	12.17± 0.71	48.73± 3.20^{a}	2.72± 0.20^{b}	2.58± 0.26	0.24± 0.02^{b}
1500 ppm Pb stress+vermicompost	2.50± 0.35	24.95± 2.26	3.50± 0.47	12.00± 1.41	26.17± 4.17^{c}	1.61± 0.24^{c}	2.64± 0.92	0.28± 0.07^{b}
LSD	1.15 ns.	3.87 ns.	0.46 ns.	2.79 ns.	8.04**	0.55**	1.41 ns.	0.13*

SV: Scale value, PH: Plant height, SD: Stem diameter, NL: Number of leaves (number plant^{-1}), PFW: Plant Fresh Weight, PDW: Plant dry weight, RFW: Root fresh weight, RDW: Root dry weight

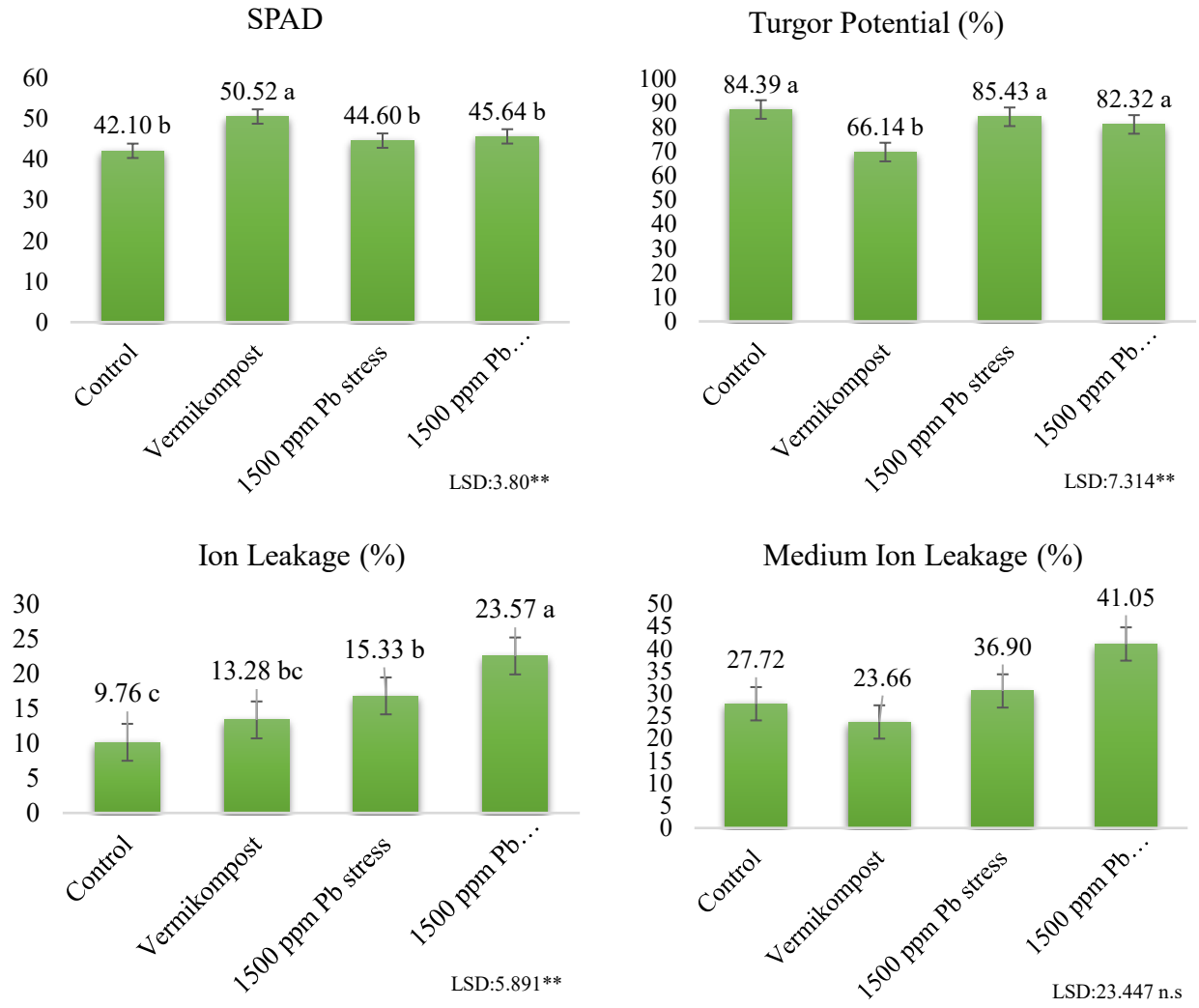


Figure 1. Effects of vermicompost, Pb stress and Pb stress+vermicompost on some physiological parameters in rocket plants

[The data followed by the same letters in the table show that there is no significant difference between the groups (LSD; Least significant difference, NS; Not significant, *p<0.05, **p<0.01,***p<0.001)]

Root Dry Weight

In terms of the root dry weight, the statistical difference between the treatments was found to be significant. Examination of root dry weight values revealed that the control treatment exhibited the highest value (0.49 g). Root dry weight decreased under vermicompost (0.27 g), 1500 ppm Pb stress (0.24 g), and Pb + vermicompost (0.28 g) applications compared with the control. The lowest root dry weight was obtained in the 1500 ppm Pb treatment. Previous research supports these findings: Pb stress generally decreases root dry weight (Balcı, 2019), and vermicompost applications have shown mixed effects. Some studies reported decreased root dry weight with vermicompost (Beykkhormizi et al. 2016), while others found variable results (Bachman and Metzger, 2007; Paul and Metzger, 2005).

The study assessed SPAD values, turgor potential, ion leakage, and medium EC for the Bengi rocket cultivar under different treatments: control, vermicompost, 1500 ppm Pb stress, and 1500 ppm Pb stress + vermicompost (Figure 1).

SPAD

Statistical analysis revealed significant differences in SPAD values among treatments. SPAD values reflecting leaf chlorophyll content varied depending on the treatments (Figure 1). The lowest SPAD value (42.10) was recorded in the control group, whereas the highest value (50.52) was obtained under the vermicompost treatment. Under 1500 ppm Pb stress, the SPAD value was measured as 44.60, indicating an increase compared with the control group. In the 1500 ppm Pb + vermicompost combination, the SPAD value was determined to be 45.64. These findings demonstrate that vermicompost application enhanced leaf chlorophyll content, while Pb stress resulted in a limited increase in SPAD values, although not to the level observed in the vermicompost-only treatment. Pb stress generally decreases SPAD values due to chlorosis and impaired photosynthesis (Balcı, 2019). Vermicompost has been reported to improve leaf chlorophyll content under stress conditions (Kıran, 2019). Despite some studies showing increased fluorescence intensity with Pb stress in other plants (Kohli et al. 2019), heavy metals typically limit photosynthesis and chlorophyll synthesis (Sharma and Dubey, 2005; Asri and Sonmez, 2006). Vermicompost, known for enhancing photosynthesis by improving plant aeration and water retention, positively affects photosynthetic parameters (Arancon et al. 2003; Levinsh, 2011). However, findings on its impact on chlorophyll content have been mixed (Berova and Karanatsidis, 2009; Ali et al. 2007).

Turgor Potential

The turgor potential was determined as 84.39% in the control group. In the vermicompost treatment, turgor potential decreased to 66.14%, representing the lowest value among the evaluated treatments. Under 1500 ppm Pb stress, turgor potential was measured at 85.43%, showing a value similar to, and slightly higher than, that of the control group. In the Pb + vermicompost combination, turgor potential was determined as 82.32%, which was close to the values observed in the control and Pb stress treatments. Overall, vermicompost application led to a pronounced reduction in turgor potential, whereas under Pb stress, turgor potential remained close to the control level. Previous studies indicate that heavy metal stress reduces relative water content (RWC) due to decreased hydraulic conductivity (Balcı, 2019; Ehlert et al. 2009), and vermicompost has been shown to improve RWC under stress (Kıran, 2019). The effect of vermicompost on RWC varies, with some studies reporting increased RWC (Verma et al. 2013), while others find no significant change (Paul and Metzger, 2005; Jain et al. 2012).

Ion Leakage

The effects of the treatments on ion leakage (%), an indicator of cell membrane integrity, are presented in Figure 1. Ion leakage values exhibited a clear increasing trend depending on the treatments. The lowest ion leakage value was recorded in the control group (9.76%). Ion leakage increased to 13.28% under the vermicompost treatment, indicating a higher value compared with the control. Under 1500 ppm Pb stress, ion leakage was measured at 15.33%, which was higher than that observed in the vermicompost treatment. The highest ion leakage value (23.57%) was obtained in the 1500 ppm Pb + vermicompost combination. The pronounced increase in ion leakage under this treatment indicates that Pb stress adversely affected cell membrane integrity and that this effect became more pronounced when Pb stress was combined with vermicompost application. Previous research supports that vermicompost can reduce electrolyte leakage under stress conditions (Kıran, 2019), and heavy metals can impair membrane permeability and water content (Costa and Morel, 1994; Kohli et al. 2019).

Medium Ion Leakage

The effects of the treatments on medium ion leakage (%) are presented in Figure 1. Although increases and decreases in the values were observed depending on the treatments, statistical analysis revealed that the differences among treatments were not significant (LSD: 23.447; $p>0.05$). The medium ion leakage value was determined as 27.72% in the control group and decreased to 23.66% under the vermicompost treatment.

Under 1500 ppm Pb stress, medium ion leakage increased to 36.90%, while the highest value (41.05%) was recorded in the 1500 ppm Pb + vermicompost combination.

Leaf micro element content

In our study, the effects of control, vermicompost, 1500 ppm lead stress, and 1500 ppm lead stress + vermicompost treatments on the micro-mineral elements Copper (Cu), Manganese (Mn), Iron (Fe), Zinc (Zn), and Lead (Pb) in Bengi rocket leaves were assessed (Figure 2). The contents of Cu, Mn, Fe, and Zn did not show statistically significant differences among treatments. However, Pb content varied significantly.

Copper content was highest in the control treatment (10.00 ppm) and lowest in the vermicompost treatment (6.33 ppm). Under 1500 ppm Pb stress, copper uptake increased with the addition of vermicompost. While vermicompost has been reported to reduce Cu content in lettuce under certain conditions, it increased Cu content under lower salt stress (4 dS m^{-1}) but decreased it under higher salt stress (8 dS m^{-1}) (Kıran, 2019).

Gutiérrez-Miceli et al. (2007) noted that vermicompost can negatively affect plant nutrient content, although its

nutrients are typically in a form readily available for plant uptake (Edwards and Burrows, 1988).

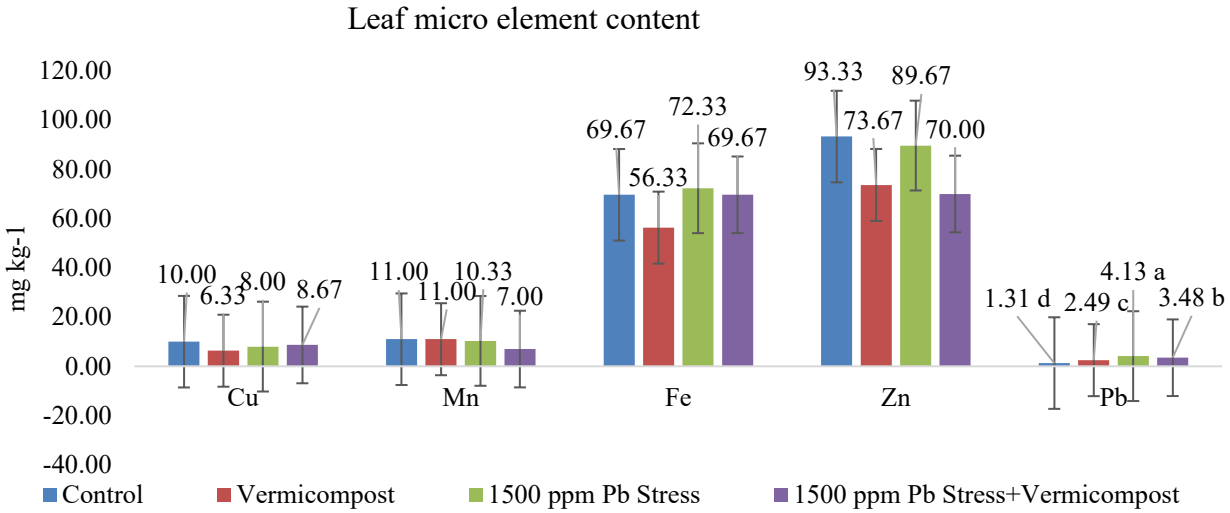
Manganese content was highest in the control and vermicompost treatments (11 ppm) and lowest in the 1500 ppm Pb stress + vermicompost treatment (7 ppm). Manganese levels were similar between the control and vermicompost treatments. Under salt stress conditions, Mn content in lettuce decreased, and vermicompost applications were ineffective in increasing Mn content (Kıran, 2019).

Iron (Fe) content was highest in the 1500 ppm Pb stress treatment (72.33 ppm) and lowest in the vermicompost treatment. Fe uptake was reduced when combining 1500 ppm Pb stress with vermicompost, resulting in 69.67 ppm Fe. Fe content was similar between the control and the 1500 ppm Pb stress + vermicompost treatment.

Previous research has shown that vermicompost applications can increase Fe content in lettuce, particularly under salt stress conditions (Kıran, 2019).

Zinc (Zn) content was highest in the control treatment (93.33 ppm) and lowest in the 1500 ppm Pb stress + vermicompost treatment (70.00 ppm). Zn levels in the control (93.33 ppm) and 1500 ppm Pb stress treatment (89.67 ppm) were similar. Vermicompost has been reported to increase Zn content in lettuce under salt stress conditions and alleviate the negative effects of salt stress on Zn content (Kıran, 2019).

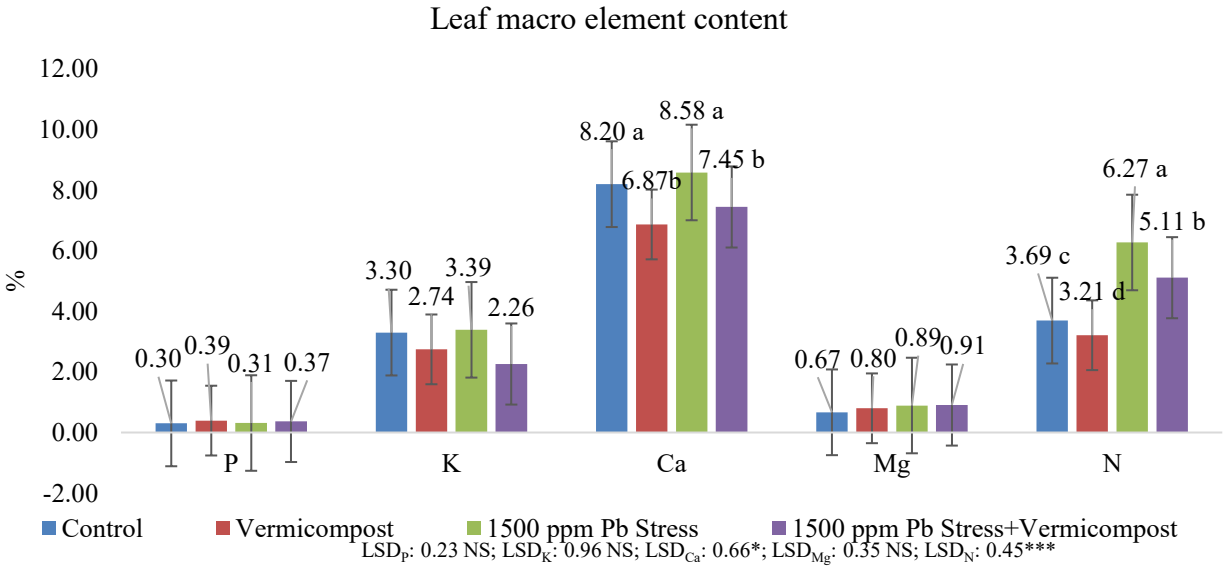
Lead (Pb) content was highest in the Pb stress treatment (4.13 ppm) and lowest in the control (1.31 ppm). Pb content was reduced in the 1500 ppm Pb stress + vermicompost treatment compared to Pb stress alone.



LSD_{Cu}: 5.6 NS; LSD_{Mn}: 7.55 NS; LSD_{Fe}: 18.46 NS; LSD_{Zn}: 32.06 NS; LSD_{Pb}: 0.49***

Figure 2. Effects of control, vermicompost, Pb stress, and Pb stress+vermicompost treatments on the microelement content in the leaves of rocket plants

[The data followed by the same letters in the table indicate that there is no significant difference between the groups (LSD; Least Significant Difference, NS; Not significant, *p<0.05, **p<0.01, *p<0.001)].



LSD_{P}: 0.23 NS; LSD_{K}: 0.96 NS; LSD_{Ca}: 0.66*; LSD_{Mg}: 0.35 NS; LSD_{N}: 0.45***

Figure 3. Effects of control, vermicompost, Pb stress, and Pb stress+vermicompost treatments on the macroelement content in the leaves of rocket plants

[The data followed by the same letters in the table show that there is no significant difference between the groups (LSD; Least significant difference, NS; Not significant, *p<0.05, **p<0.01,***p<0.001)]

Previous studies have reported similar findings, with a 26% reduction in Pb uptake when using Sonchus aqueous extract in wheat (Nessem et al. 2023). Pb content in rocket leaves has been reported as 0.93 ppm in control and 2.16 ppm in 1500 mg kg^{-1} Pb treatment (Yıldırım et al. 2019).

Leaf macro element content

The effects of various treatments on the macro-element content of phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), and nitrogen (N) in Bengi rocket leaves are presented in Figure 3.

Phosphorus (P) content was highest in the vermicompost treatment (0.39%) and lowest in the control treatment (0.30%). Although phosphorus content increased with the combination of 1500 ppm Pb stress and vermicompost compared to 1500 ppm Pb stress alone, the differences among treatments were not statistically significant. Previous studies have noted that vermicompost can enhance phosphorus content in plants, especially under salt stress conditions (Kıran, 2019). However, a negative correlation between phosphorus uptake and soil lead concentration has also been reported (Akıncı et al. 2010; Päivöke, 2002).

Potassium (K) content did not show significant differences among treatments overall. The highest potassium content (3.39%) was observed in the 1500 ppm Pb stress treatment, while the lowest was found in the 1500 ppm Pb stress + vermicompost treatment. Vermicompost applications were reported to increase potassium content in bean leaves compared to controls (Beykkhormizi et al. 2016), and to enhance potassium content under salt stress conditions (Kıran, 2019). Lead exposure was associated with reduced potassium content in plants, possibly due to competition between lead and potassium ions for uptake through similar channels (Wang et al. 2008; Pourrut et al. 2008; Sharma and Dubey, 2005). Significant decreases in potassium content have been reported in plants under lead stress (Päivöke, 2002; Akıncı et al. 2010).

The effects of different treatments on the macro-element content of calcium (Ca), magnesium (Mg), and nitrogen (N) in Bengi rocket leaves are summarized as follows:

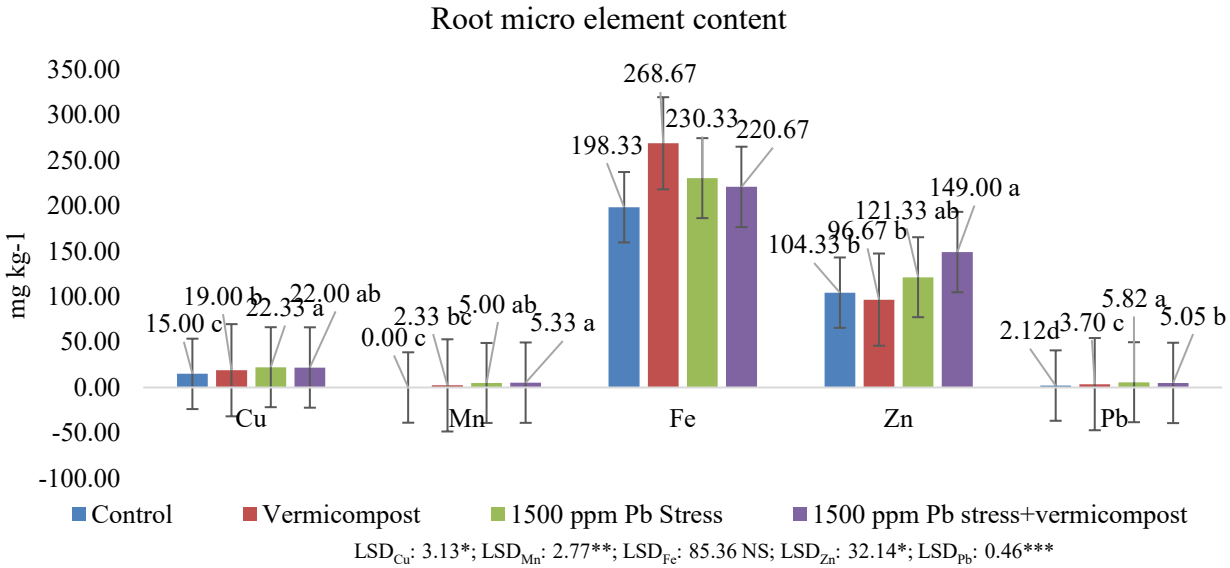
Calcium (Ca) Content: Significant differences were observed among treatments. Calcium content was highest in the 1500 ppm Pb stress treatment (8.58%) and lowest in the vermicompost treatment. The presence of vermicompost reduced Ca uptake in the 1500 ppm Pb stress treatment, with Ca content decreasing to 7.45% in the 1500 ppm Pb stress + vermicompost treatment. Lead's similarity to Ca allows it to displace Ca^{2+} in plant cells, disrupting calcium uptake (Azmat et al. 2009). Previous studies have also reported that high lead concentrations can reduce the uptake of macro elements like Ca in plants (Lamhamdi et al. 2013).

Magnesium (Mg) Content: There were no statistically significant differences among treatments for magnesium content, but it was lowest in the control treatment and highest in the 1500 ppm Pb stress + vermicompost treatment. Vermicompost increased Mg content under Pb stress conditions. Lead stress negatively impacts Mg levels, with significant reductions observed even at low concentrations, due to lead's interference with chlorophyll synthesis and Mg uptake (Sengar and Pandey, 1996; Haider et al. 2006).

Nitrogen (N) Content: Significant differences were found in nitrogen content among treatments. The highest N content was observed in the 1500 ppm Pb stress treatment. The addition of vermicompost reduced N content in both Pb stress conditions and the vermicompost-only treatment, with the lowest N content (3.21%) in the vermicompost treatment.

Lead toxicity often leads to reduced photosynthesis and disruptions in chloroplast function, affecting N content through various mechanisms, including the inhibition of chlorophyll synthesis and alteration of protein and enzyme activities (Xiong et al. 2006; Gupta et al. 2009; Cenkci et al. 2010).

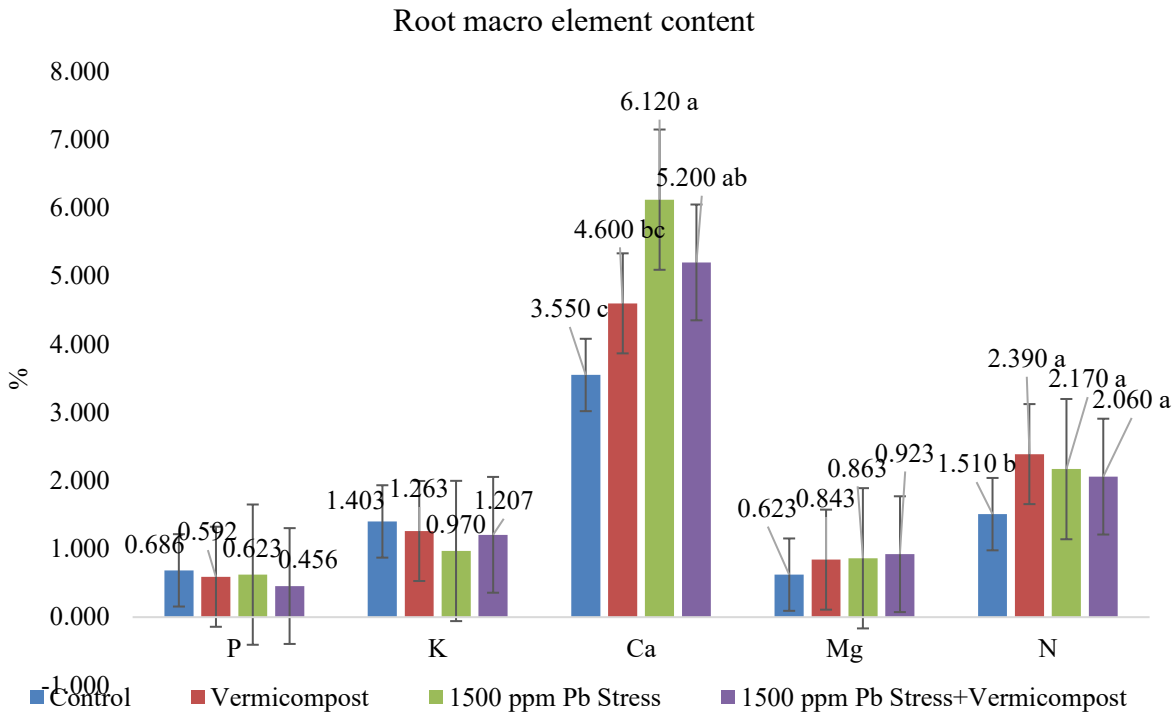
Overall, lead stress impacts the uptake and content of macro-elements differently, with vermicompost showing varied effects on these parameters under stress conditions.



LSD_{Cu}: 3.13*; LSD_{Mn}: 2.77**; LSD_{Fe}: 85.36 NS; LSD_{Zn}: 32.14*; LSD_{Pb}: 0.46***

Figure 4. Effects of control, vermicompost, Pb stress, and Pb stress+vermicompost treatments on the microelement content in the roots of rocket plants

[The data followed by the same letters in the table show that there is no significant difference between the groups (LSD; Least significant difference, NS; Not significant, *p<0.05, **p<0.01,***p<0.001)]



LSD_P: 0.43 NS; LSD_K: 0.47 NS; LSD_{Ca}: 1.33*; LSD_{Mg}: 0.3 NS; LSD_N: 0.51*

Figure 5. Effects of control, vermicompost, Pb stress, and Pb stress+vermicompost treatments on the macroelement content in the roots of rocket plants

[The data followed by the same letters in the table show that there is no significant difference between the groups (LSD; Least significant difference, NS; Not significant, *p<0.05, **p<0.01,***p<0.001)]

Root micro element content

The effects of various treatments on the micro-mineral element content (Figure 4). in Bengi rocket roots are summarized as follows:

Copper (Cu) Content: Significant differences were observed among treatments. Root Cu content was lowest in the control treatment (15 ppm) and highest in the 1500 ppm Pb stress treatment (22.33 ppm). Cu content decreased in the 1500 ppm Pb stress + vermicompost treatment compared to the Pb stress alone. All treatments other than the control resulted in a lower Cu content.

Manganese (Mn) Content: Statistically significant differences were found in root Mn content. The highest Mn content was observed in the 1500 ppm Pb stress + vermicompost treatment (5.33 ppm), while the control treatment showed the lowest Mn content (0.00 ppm). Mn content increased with the addition of vermicompost under Pb stress conditions, demonstrating an enhancement compared to Pb stress alone.

Iron (Fe) Content: No statistically significant differences were noted among treatments. The highest Fe content was recorded in the vermicompost treatment (268.67 ppm). In the 1500 ppm Pb stress + vermicompost treatment, Fe content decreased. Overall, Fe content was higher in all treatments compared to the control.

Zinc (Zn) Content: Significant differences were observed in root Zn content. Zn levels were 96.67 ppm in vermicompost, 104.33 ppm in control, 121.33 ppm in 1500 ppm Pb stress, and 149.00 ppm in 1500 ppm Pb stress + vermicompost treatment. Zn content increased with Pb stress and further increased in the presence of vermicompost, compared to the control and other treatments.

Lead (Pb) Content: Differences in Pb content among control, vermicompost, 1500 ppm Pb stress, and 1500 ppm Pb stress + vermicompost treatments were not statistically significant. Pb content was highest in the 1500 ppm Pb stress treatment and lowest in the control treatment. Pb content was 3.70 ppm in the vermicompost treatment. The addition of vermicompost in the 1500 ppm Pb stress treatment reduced Pb content compared to Pb stress alone. The application of vermicompost with Pb stress effectively lowered Pb content in rocket roots.

In summary, Cu and Mn contents showed significant variations across treatments, with the highest Mn content associated with Pb stress + vermicompost. Fe content did not vary significantly among treatments but was highest with vermicompost. Zn content increased in Pb stress treatments and further increased with vermicompost.

Root Macro Element Content

The effects of various treatments on the macro-mineral element content (Figure 5) in Bengi rocket roots are summarized below:

Phosphorus (P) Content: No statistically significant differences were observed among the treatments.. The highest root P content was found in the control treatment (1.403%), followed by vermicompost (1.263%), 1500 ppm Pb stress + vermicompost (1.207%), and 1500 ppm Pb stress (0.970%). The presence of vermicompost under Pb stress decreased P content in the roots.

Calcium (Ca) Content: Significant differences were noted.

The highest Ca content was recorded in the 1500 ppm Pb stress treatment. The addition of vermicompost reduced Ca content in the 1500 ppm Pb stress treatment. In contrast, Ca content increased with vermicompost application compared to the control. Previous studies have shown varying effects of vermicompost on Ca content, with some reports indicating increased Ca in bean roots under certain vermicompost applications.

Magnesium (Mg) Content: No statistically significant differences were observed among the treatments.. The highest Mg content was found in the 1500 ppm Pb stress + vermicompost treatment (0.923%), while the lowest was in the control treatment (0.623%). Mg content increased with vermicompost application, particularly under Pb stress conditions.

Potassium (K) Content: No statistically significant differences were observed among the treatments.. The lowest K content was observed in the 1500 ppm Pb stress treatment, while vermicompost application increased K content in the 1500 ppm Pb stress treatment. Previous studies have indicated that vermicompost can increase K content, although effects can vary with application rates.

Nitrogen (N) Content: Statistically significant differences were found. The highest N content was in the vermicompost treatment (2.390%), while the lowest was in the control treatment (1.510%). The presence of vermicompost in the 1500 ppm Pb stress treatment decreased N content. While some studies report increased total nitrogen content with vermicompost, others find no significant effect.

In summary, the addition of vermicompost had variable effects on mineral element contents in rocket roots, showing notable improvements in Mg and N content but reducing Ca and P content under Pb stress conditions.

Conclusion

This study investigated the effects of vermicompost on the growth, physiological traits, membrane stability, and mineral nutrient composition of Bengi rocket (Eruca sativa L.) under 1500 ppm Pb stress. Pb stress significantly reduced growth parameters, including leaf number and plant and root biomass. Vermicompost increased SPAD values, indicating improved chlorophyll content, but did not enhance growth under Pb stress. Ion leakage was highest in the Pb + vermicompost treatment, suggesting increased membrane damage. Although vermicompost reduced Pb accumulation in leaves and roots and promoted the uptake of some nutrients, particularly Mg and N in roots, these effects were insufficient to alleviate the negative impacts of severe Pb stress. Overall, vermicompost showed limited effectiveness in improving Pb tolerance under the conditions tested. Further studies are needed to evaluate different vermicompost doses and Pb levels to optimize its potential use in heavy metal stress management.

Declarations

Ethical Approval Certificate

Ethics committee approval is not required.

Author Contribution Statement

Conceptualization, *Sultan Dere Uğur*.; methodology, *Sultan Dere Uğur*.; software, *Sultan Dere Uğur*.; validation, *Sultan Dere Uğur*.; formal analysis, *Sultan Dere Uğur*.; investigation, *Sultan Dere Uğur*.; resources, *Sultan Dere Uğur*.; data curation, *Sultan Dere Uğur*.; writing-original draft preparation, *Sultan Dere Uğur*.; writing-review and editing, *Sultan Dere Uğur*.; visualization, *Sultan Dere Uğur*.; supervision, *Sultan Dere Uğur*.; project administration, *Sultan Dere Uğur*..; and funding acquisition, *Sultan Dere Uğur* All authors have read and agreed to the published version of the manuscript.

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Conflict of Interest

The authors declare no conflict of interest.

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