

# JOURNAL OF WATER AND LAND DEVELOPMENT

e-ISSN 2083-4535



Polish Academy of Sciences (PAN)

Institute of Technology and Life Sciences - National Research Institute (ITP - PIB)

JOURNAL OF WATER AND LAND DEVELOPMENT DOI: 10.24425/jwld.2025.155309 2025, No. 66 (VII–IX): 128–136

# The effect of zeolite on yield of *Lactuca sativa* var. *capitata* and chemical composition

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RECEIVED 26.03.2025

ACCEPTED 23.06.2025

AVAILABLE ONLINE 17.09.2025

**Abstract:** This study evaluated the impact of different doses of zeolite on the emergence, yield, and chemical composition of lettuce seedlings (*Lactuca sativa* var. *capitata*). To achieve this, a pot experiment was conducted using randomised design with four replications. Zeolite was mixed with the soil substrate at rates of 1%, 5%, and 10% per 1 kg of total soil mass (5 kg), corresponding to 10, 50, and 100 g·kg<sup>-1</sup>, respectively. The control treatment consisted of a light mineral soil substrate without the addition of zeolite.

The study found that zeolite significantly improved lettuce seed germination and seedling emergence rate. As the proportion of zeolite in the soil increased, both the yield of edible parts (leaves) and the biomass of the roots also increased. A beneficial effect of zeolite was observed to significantly reduce cadmium and lead concentrations in leaves. Furthermore, the addition of 10 g·kg<sup>-1</sup> of soil also led to a reduction in the nitrate content in leaves, while higher doses (50 and 100 g·kg<sup>-1</sup> of substrate) resulted in an increased nitrate content. The application of zeolite had no effect on the levels of flavonoids, anthocyanins, or polyphenols in lettuce leaves, nor on the concentrations of primary nutrients (N, P, K, Ca, Mg, S, and Na). However, a statistically significant relationship was observed between the application of zeolite and the increase in the iron, copper and zinc content, along with the decrease in the levels of manganese, boron, and molybdenum in the edible parts of the test plant.

Keywords: leaf yield, lettuce, nutrient content, root yield, seedling emergence

# INTRODUCTION

The ongoing global climate changes is expected to have particularly severe impacts on agriculture, affecting not only crop yields, but also their adaptability to the prevailing environmental conditions. In some regions, extreme weather events – such as heatwaves and droughts – are already adversely impacting agricultural production. This leads to economic losses, primarily due to water shortages, which in turn result in nutrient imbalances. Such imbalances are particularly concerning when

they involve elements that can have adverse effects on humans and animals (Mondal et al., 2021).

An important adaptive strategy to mitigate the effects of climate change in agriculture involves reducing water use, including the adoption of water-efficient technologies. The efficiency of nutrient uptake and plant growth is closely related to the physical and chemical properties of the soil (Lipiński, Lipińska and Shuvar, 2021). Therefore, the application of soil amendments, especially natural or organic ones, plays a significant role in the long-term improvement of soil physicochemical

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properties (Jarosz *et al.*, 2022). In this context, the use of zeolite is an interesting solution; however, its application in Poland remains limited (Kavvadias *et al.*, 2023). The effects of zeolite use vary depending on factors such as its type, dosage, particle size and density, application method, soil texture and structure, as well as moisture and salinity conditions (Nakhli *et al.*, 2017).

Zeolites are a group of aluminosilicate minerals with diverse structures, chemical compositions, and crystalline forms. They are widely used in various industries, including agriculture and horticulture (Sangeetha and Baskar, 2016; Eroglu, Emekci and Athanassiou, 2017). Their properties, particularly porosity and cation exchange capacity, are critical for plant nutrition. Furthermore, their ability to selectively bind and slowly release ions into soil solution makes them particularly valuable for optimised fertilisation strategies (Kavvadias *et al.*, 2023). Additionally, zeolites enhance phosphorus solubility, thereby improving its bioavailability to plants (Sangeetha and Baskar, 2016).

A particularly important characteristic of zeolite in horticultural production is its ability to mitigate salt stress. This justifies its application in salt-sensitive crops, as well as in cases of high doses of fertilisers or drought conditions. Additionally, zeolites can also modify soil water retention by altering its bulk density and porosity (Nakhli *et al.*, 2017).

Numerous studies have demonstrated the effectiveness of the application of zeolite, including in vegetable production (Sangeetha and Baskar, 2016; Türkmen and Su, 2019; Babaousmail et al., 2022; Kavvadias et al., 2023; Conversa et al., 2024; Santos, Shinzato and Castanho, 2024). Consequently, the present study investigates the impact of zeolite on lettuce cultivation under Polish conditions. In Polish legal regulations, the category of plant growth support agents includes zeolite, which meets the definition of a soil-improving agent. It is not classified as a fertiliser product, but rather as a substance added to the soil to enhance its chemical, physical, physicochemical, or biological properties. Zeolite may also meet the criteria for mineral growth stimulants, which positively influence plant development (Ustawa, 2007).

The objective of this study was to evaluate the effect of zeolite mixed with soil substrate on early growth, yield, and chemical composition of the test plant under controlled pot experiment conditions. It was hypothesised that the addition of zeolite to the soil would have a positive impact on the studied parameters, but the effects would vary depending on its percentage share in the substrate.

### **MATERIALS AND METHODS**

The effect of zeolite on lettuce (*Lactuca sativa* var. *capitata*) was evaluated in a pot experiment carried out at the Didactic and Research Station in Sosnowica during 2022–2023. The experiment followed a completely randomised design, with four replications and two series per year. The pots were filled with 5 kg of soil mixed with zeolite at different doses: 1, 5 and 10% of the soil mass, corresponding to 10, 50, and 100 g·kg<sup>-1</sup> of substrate, respectively. Control treatment consisted of soil without addition of zeolite. The study used light mineral soil with a pH of 5.89, characterised by: medium availability of phosphorus (14.3 mg P<sub>2</sub>O<sub>5</sub> per 100 g of soil), low potassium content (8 mg K<sub>2</sub>O per 100 g of soil) and medium magnesium

content (4.8 mg Mg per 100 g of soil). The applied zeolite had the following characteristics:

- pH in KCl: 5.2;
- cation exchange capacity (mmol·kg<sup>-1</sup>): 206.8;
- granulometric composition:
- 0-2.0 mm: 0.28%,
- 5-0.99 mm: 89.66%,
- 25-0.49 mm: 9.89%,
- <0.25 mm: 0.17%.

The size of the dominant particles, which accounted for 99.6%, was 0.25–0.99 mm. The heavy metal content in the zeolite is presented in Table 1.

**Table 1.** The heavy metal content in zeolite was analysed in comparison to the legal requirements for fertilisers and plant growth support products (PGSP) (Rozporządzenie, 2024)

Metal	Content (mg·kg <sup>-1</sup> )	Permissible contents in mg per 1 kg of mineral fertiliser or PGSP
Fe	56,320	no acceptable threshold values defined
Zn	340	no acceptable threshold values defined
Cu	490	no acceptable threshold values defined
Mn	510	no acceptable threshold values defined
Pb	90	140
Cd <sup>1)</sup>	-	5
Hg <sup>1)</sup>	_	2

Below the detection limit. Source: own elaboration.

A constant fertilisation regime was applied throughout the experiment: nitrogen (N): 0.25 mg·kg<sup>-1</sup> of soil (as ammonium nitrate), phosphorus (P): 0.3 g·kg<sup>-1</sup> (as triple superphosphate) and potassium (K): 0.9 g·kg<sup>-1</sup> (as potassium chloride). Lettuce seeds (30 per pot) were sown twice each year: first series in late March and second series in mid-July. Seed germination was evaluated seven days after sowing, defined as the emergence of embryonic roots at least as long as the seed or its diameter (ISTA, 2012). Furthermore, the number of normal seedlings, abnormal seedlings, and dead seeds were recorded. Abnormal seedlings and dead seeds were removed, and germination results were expressed as percentages. Each pot was then thinned to maintain an equal number of plants. The remaining seedlings were grown until harvest maturity (10-12 cm in height). Five weeks after sowing, the aboveground parts (leaves) were harvested early in the morning to ensure maximum freshest. Root systems were carefully collected by rinsing with distilled water over a plastic sieve. Fresh plant material (leaves and roots) was weighed to determine fresh yield (g per pot), after which the samples were subjected to chemical analysis. Fresh lettuce leaves were washed with distilled water and dried at 105°C for three hours in a forcedair laboratory oven (30 L PRO). The dried samples were ground using a FRITSCH Pulverisette 14 rotary mill (Idar-Oberstein, Germany) with an adjustable rotation speed. To determine the macronutrient and micronutrient contents (N, K, P, Ca, Mg, S, Na, B, Cu, Fe, Mn, Mo, and Zn), as well as heavy metals (Cd, Pb), plant samples were mineralised in acid solutions. Nitrogen (N) was measured using the Kjeldahl method (Persson and Wennerholm, 1999), while the remaining elements were analysed using inductively coupled plasma optical emission spectrometry (ICP-OES) with a Shimadzu ICPE-9800 system (Shimadzu, Japan). Nitrate content was determined by continuous flow analysis (CFA) with spectrophotometric detection (according to PN-EN 12014-7:2001). In compliance with legal regulations, the contents of nitrate, cadmium, and lead were expressed relative to fresh mass, while all other elements were reported on a dry mass basis. Additionally, cadmium and lead concentrations were also determined in lettuce roots.

To determine the total polyphenol content in lettuce leaves, 1 g of each sample was extracted with 3 cm³ of methanol at room temperature using a shaker. The extracts were centrifuged, and the solid residue was extracted twice using methanol (2·3 cm³). All three methanol extracts were combined and the final volume was adjusted to 10 cm³. The total polyphenol content was determined using the Folin-Ciocalteu (FC) reagent, following Yu et al. (2005). The reaction mixture contained 250 mm³ of fresh FC reagent, 750 mm³ of 20% Na<sub>2</sub>CO<sub>3</sub>, and 3 cm³ of deionised water, added to 50 mm³ of sample solution. After two hours of reaction at room temperature, absorbance was measured at 765 nm.

To determine the flavonoid content, 0.5 g of lettuce leaves were extracted with a mixture of acetone and HCl under reflux boiling conditions. After cooling and filtering, the extract was further extracted with ethyl acetate. Aluminium chloride solution was added to the extract, forming yellow-coloured flavonoid-aluminium complexes. The absorbance of the complexes was measured at  $\lambda_{\rm max}=425$  nm.

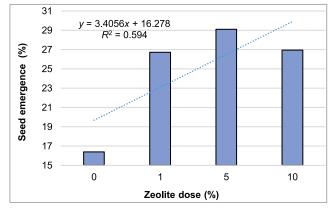
To obtain a homogeneous analytical sample for the determination of anthocyanins in lettuce leaves, the plant material was ground while frozen in a blender using dry ice (solid CO<sub>2</sub>). Then, 10 g of the ground sample were extracted with 50 cm<sup>3</sup> of 70% methanol for 2 min. The extract was centrifuged (10,000 rpm, 10 min), filtered, and diluted (1:3) in 70% methanol. 5 mm<sup>3</sup> of the extract were analysed using high performance liquid chromatography (HPLC) with a diode array detector (DAD). Chromatographic separation was performed on a Phenomenex® Fusion-RP 80A column (250 mm×4.6 mm; 4 µm) with a precolumn. The elution conditions were as follows: flow rate 1 cm<sup>3</sup>·min<sup>-1</sup>, temperature 25°C, wavelength 520 nm. The mobile phase consisted of 5% formic acid (solvent A) and acetonitrile (solvent B). The HPLC analysis was performed in gradient flow: 0-16 min: 3-9% B (linear), 16-30 min: 12% B (linear), 30-54 min: 33% B (linear), 54-58 min: 90% B (linear), 58-62 min: 90% B (isocratic), 62-64 min: 3% B (linear), 64-65 min: 3% B (isocratic). The HPLC column was then re-equilibrated to the initial gradient conditions for 15 min.

The collected data were subjected to one-way analysis of variance (ANOVA) to evaluate the effect of different doses of the tested zeolite on the content of macroelements, microelements, and bioactive compounds in plant leaves. Prior to the analysis, the assumptions of normality and homogeneity of variance were verified. When the ANOVA indicated statistically significant differences (p < 0.05), Tukey's honest significant difference (Tukey HSD) post hoc test was applied to determine which treatment means differed significantly from each other, as well as correlation and linear regression methods. All statistical analyses were performed at a significance level of  $\alpha = 0.05$ .

# **RESULTS**

# SEEDLING EMERGENCE

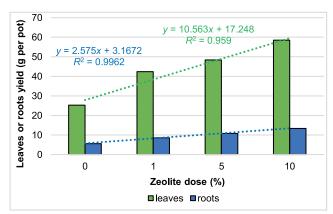
The application of zeolite at 1, 5, and 10% as an amendment to the soil substrate in which lettuce was grown resulted in significant differences in seedling emergence compared to the control treatment, particularly in cases where 1 and 5% zeolite was applied (Fig. 1). In these two treatments, seedling emergence was more than 10% higher than in the control group. The highest emergence rate was recorded with the addition of 5% zeolite, reaching 29.11%. No significant differences were observed between the emergence rates between 1 and 10% zeolite treatments.



**Fig. 1.** Lettuce seedling emergence (%) under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); significant differences between treatments:  $0^a$ ,  $1^b$ ,  $5^b$ ,  $10^b$  (p < 0.001); letter groupings indicate statistically significant differences between doses (at a significance level of p < 0.05); source: own study

# **DRY MASS OF PLANTS**

The application of zeolite significantly increased lettuce yield, regardless of the dosage added to the soil substrate (Fig. 2). The highest yield of edible parts was observed at the highest dose of zeolite (10%). The fresh mass of leaves more than doubled



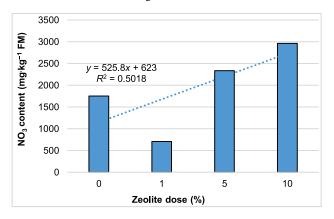
**Fig. 2.** Yields of fresh weight of lettuce leaves and roots under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); significant differences in leaf biomass between treatments:  $0^a$ ,  $1^{ab}$ ,  $5^b$ ,  $10^b$  (p < 0.001), and in root biomass:  $0^a$ ,  $1^{ab}$ ,  $5^{ab}$ ,  $10^b$  (p = 0.048); letter groupings indicate statistically significant differences between doses (at a significance level of p < 0.05); source: own study

compared to the control treatment and increased by 20 to 40% with 1 and 5% addition of zeolite, respectively. A similar trend was observed for root biomass, with the increases being even more pronounced than those observed in the aboveground parts (Fig. 2).

### CHEMICAL COMPOSITION OF PLANTS

### Nitrate content

Nitrate content is one of the key quality indicators for lettuce. The addition of zeolite had a significant effect on the  $NO_3$  concentration in the edible parts of the plants. In the 1% zeolite treatment, the nitrate content in lettuce leaves was three times lower than in the fresh mass of leaves from the control group. However, a fivefold and tenfold increase in the zeolite dose resulted in a gradual rise in nitrate levels (Fig. 3). Importantly, all recorded nitrate concentrations remained well within the legal quality standards for lettuce, which set a limit of 3,000 mg·kg<sup>-1</sup> fresh mass (Commission Regulation, 2011).



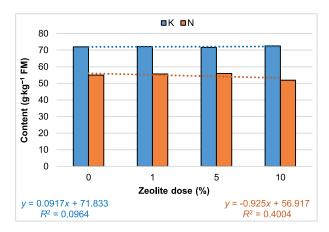
**Fig. 3.** Content of NO<sub>3</sub> in lettuce leaves under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); FM = fresh mass; significant differences between treatments:  $0^b$ ,  $1^a$ ,  $5^c$ ,  $10^d$  (p < 0.001); letter groupings indicate statistically significant differences between doses (at a significance level of p < 0.05); source: own study

# Macro- and micronutrient content

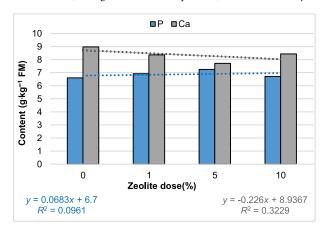
The addition of zeolite to the soil had limited effect on the macronutrient content in lettuce leaves (Figs. 4–6). Nitrogen and potassium levels remained generally similar across most treatments, with the exception of potassium levels under the application of zeolite at 5% of substrate biomass, where significantly lower values were observed compared to the other treatments. In contrast, calcium and phosphorus contents showed a slight decrease with increasing zeolite dosage.

Statistically significant differences in the content of these macronutrients were recorded only in the treatment with 5% zeolite addition. Compared to the control, phosphorus content increased, whereas calcium content decreased. No major differences were observed in the sulphur and sodium contents. However, magnesium levels were higher compared to plant material from the control treatment, with statistically significant differences found in the treatments with lower zeolite doses (1 and 5%).

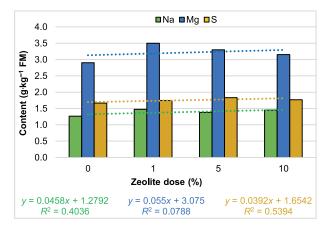
A slightly greater variation was observed in the micronutrient content, particularly for copper, iron, zinc, boron, and molybdenum (Figs. 7–9). Compared to the control treatment, the



**Fig. 4.** Nitrogen (N) and potassium (K) content in lettuce leaves under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); FM = fresh mass; significant differences in N content between treatments:  $0^a$ ,  $1^a$ ,  $5^a$ ,  $10^b$  (p < 0.001), and in K content:  $0^b$ ,  $1^b$ ,  $5^a$ ,  $10^b$  (p = 0.066); letter groupings indicate statistically significant differences between doses (at a significance level of p < 0.05); source: own study



**Fig. 5.** Phosphorus (P) and calcium (Ca) content in lettuce leaves under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); FM = fresh mass; significant differences in P content between treatments:  $0^a$ ,  $1^a$ ,  $5^b$ ,  $10^a$  (p=0.009), and in Ca content:  $0^b$ ,  $1^b$ ,  $5^a$ ,  $10^b$  (p=0.159); letter groupings indicate statistically significant differences between doses (at a significance level of p<0.05); source: own study



**Fig. 6.** Sodium (Na), magnesium (Mg) and sulphur (S) content in lettuce leaves under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); FM = fresh mass; significant differences in Na content between treatments:  $0^a$ ,  $1^a$ ,  $5^a$ ,  $10^a$  (p = 0.557), in Mg content:  $0^a$ ,  $1^b$ ,  $5^b$ ,  $10^a$  (p < 0.001), and in S content:  $0^a$ ,  $1^a$ ,  $5^a$ ,  $10^a$ ; letter groupings indicate statistically significant differences between doses (at a significance level of p < 0.05); source: own study

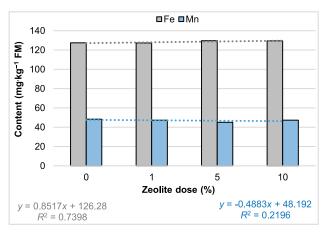
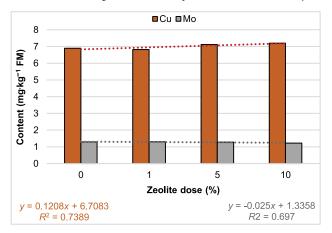
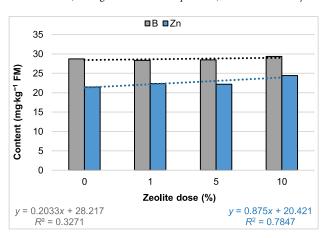


Fig. 7. Iron (Fe) and manganese (Mn) in lettuce leaves under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); FM = fresh mass; significant differences in Fe content between treatments:  $0^a$ ,  $1^a$ ,  $5^b$ ,  $10^b$  (p = 0.002) and in Mn content:  $0^a$ ,  $1^a$ ,  $5^b$ ,  $10^a$  (p < 0.001); letter groupings indicate statistically significant differences between doses (at a significance level of p < 0.05); source: own study



**Fig. 8.** Copper (Cu) and molybdenum (Mo) content in lettuce leaves under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); FM = fresh mass; significant differences in Cu content between treatments:  $0^a$ ,  $1^a$ ,  $5^b$ ,  $10^a$  (p=0.009), and in Ca content:  $0^a$ ,  $1^a$ ,  $5^a$ ,  $10^a$  (p=0.159); letter groupings indicate statistically significant differences between doses (at a significance level of p<0.05); source: own study

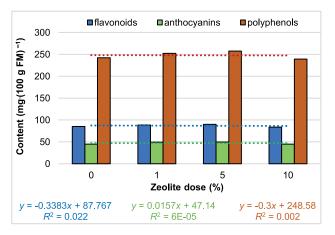


**Fig. 9.** Boron (B) and zinc (Zn) content in lettuce leaves under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); FM = fresh mass; significant differences in B content between treatments:  $0^a$ ,  $1^a$ ,  $5^a$ ,  $10^b$  (p < 0.001), and in Zn content:  $0^a$ ,  $1^a$ ,  $5^a$ ,  $10^b$  (p < 0.001); letter groupings indicate statistically significant differences between doses (at a significance level of p < 0.05); source: own study

application of zeolite significantly increased the concentration of copper, iron, zinc, and boron, while molybdenum and manganese levels showed a decreasing trend.

# Polyphenol, flavonoid and anthocyanin content

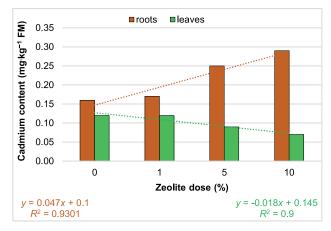
The study indicated that zeolite addition at 1% and 5% stimulated the accumulation of polyphenols, flavonoids, and anthocyanins (Fig. 10), although a statistically significant increase was observed only for anthocyanins. In contrast, the highest zeolite dose (10%) significantly reduced the levels of all these groups of compounds compared to their content in leaves from the control treatments. The only exception was anthocyanins which remained unaffected.



**Fig. 10.** Content of polyphenols, flavonoids and anthocyanins in lettuce leaves under the influence of different zeolite doses (1, 5, 10%) and the control treatment (0 – without zeolite); FM = fresh mass; significant differences in content polyphenols between treatments:  $0^b$ ,  $1^b$ ,  $5^b$ ,  $10^a$  (p = 0.018), and in flavonoids content:  $0^b$ ,  $1^b$ ,  $5^b$ ,  $10^a$  (p = 0.016) and in anthocyanins content:  $0^a$ ,  $1^a$ ,  $5^b$ ,  $10^a$  (p = 0.006); letter groupings indicate statistically significant differences between doses (at a significance level of p < 0.05); source: own study

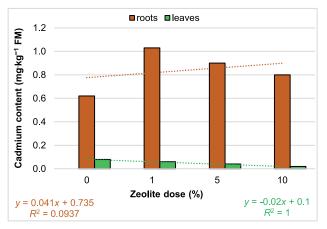
### Heavy metal content

The application of zeolite had favourable effect in reducing the heavy metal content in lettuce (Fig. 11). In the control treatment, the cadmium content in leaves reached 0.12 mg·kg<sup>-1</sup> fresh weight,



**Fig. 11.** Cadmium content in lettuce leaves and roots under the influence of different zeolite doses (1, 5, 10%) and in the control treatment (0 – without zeolite); FM = fresh mass; significant differences in Cd content in leaves:  $0^a$ ,  $1^b$ ,  $5^b$ ,  $10^b$  (p < 0.001) and in roots:  $0^a$ ,  $1^a$ ,  $5^a$ ,  $10^b$  (p = 0.002); letter groupings indicate statistically significant differences between treatments (at the p < 0.05 level); source: own study

exceeding the permissible level of 0.1 mg·kg<sup>-1</sup> fresh weight (Commission Regulation, 2021b). The addition of zeolite at a 1% dose did not reduce Cd content, whereas the 5% and 10% doses resulted in significant decreases to 0.09 and 0.07 mg·kg<sup>-1</sup> fresh weight, respectively. However, cadmium was not eliminated from the roots. In this case, cadmium levels increased progressively with higher zeolite doses. In the edible parts of lettuce (leaves), zeolite application also led to a reduction in lead content compared to the control (Fig. 12), reaching levels of 1.03, 0.085, and 0.073 mg·kg<sup>-1</sup> fresh weight, respectively, while the permissible lead content is 0.3 mg·kg<sup>-1</sup> fresh weight (Commission Regulation, 2021a).



**Fig. 12.** Lead content in lettuce leaves and roots under the influence of different zeolite doses (1, 5, 10%) and in the control treatment (0 – without zeolite); FM = fresh mass; significant differences in Pb content in leaves:  $0^{\rm c}$ ,  $1^{\rm bc}$ ,  $5^{\rm ab}$ ,  $10^{\rm a}$  (p < 0.001) and in roots:  $0^{\rm a}$ ,  $1^{\rm c}$ ,  $5^{\rm bc}$ ,  $10^{\rm ab}$  (p < 0.001); letter groupings indicate statistically significant differences between treatments (at the p < 0.05 level); source: own study

# **DISCUSSION**

Lettuce (*Lactuca sativa* L.) requires careful attention and the provision of suitable growing conditions (Ćurčić *et al.*, 2022). Therefore, zeolite – used as an inert material mixed with soil – was hypothesised to have a positive effect on seed germination, early seedling growth, dry mass accumulation, nutrient content, nutrient uptake, and overall nutrient use efficiency (Cadar *et al.*, 2022; Jawad *et.al.*, 2023).

In the present study, all applied doses of zeolite (1, 5, and 10% of substrate mass) had a significantly positive effect on seed germination and early seedling growth of lettuce. The most favourable results were observed in the treatment with a 5% zeolite addition (i.e. 5 g·kg<sup>-1</sup> of soil); however, these differences were not statistically confirmed. The positive effect of zeolite on seed germination has also been reported by Türkmen and Su (2019). Similarly, studies on other vegetable species have shown higher growth rates of lettuce seedlings in zeolite-treated soils compared to control conditions. It was demonstrated that the addition of zeolite to the soil significantly improved growth parameters such as leaf area, plant height, fruit number, and tomato biomass. Notably, the lowest zeolite dose (0.75%) yielded the best results, increasing chlorophyll content and enhancing physiological traits of the studied species (Jawad et al., 2023).

The study revealed that fresh mass yield of lettuce was significantly higher in all zeolite-treated groups compared to the control. The positive effect of zeolite on vegetable crop yield, including lettuce, has also been demonstrated by Kavvadias et al. (2023), who reported that dry mass increases of several dozen percent. Likewise, in cucumber cultivation, zeolite-treated seedlings exhibited higher biomass than those in the control (Türkmen and Su, 2019). Babaousmail et al. (2022) explains the positive effect on lettuce growth in the zeolite treatments by the fact that it can significantly reduce the stress caused by substrate salinity. This stress reduction improves plant growth and development, increases number of leaves and their area, as well as translates into higher yields of fresh and dry matter. Furthermore, Kavvadias et al. (2023) argued that zeolite (clinoptilolite) combined with fertilisers could produce effects similar to those of growth inhibitors, also reported that fertiliser effects were statistically comparable to those of clinoptilolite zeolite treatments.

The study also revealed significant differences in fresh mass accumulation between treatments with different zeolite doses. Fresh leaf and root mass increased with increasing zeolite application. In contrast, Kavvadias *et al.* (2023) found that a 2% zeolite addition significantly increased lettuce yield not only compared to the control (0% zeolite) but also compared to the 5% zeolite treatment.

While most zeolites have beneficial effects on plant growth and yield, there are instances where their effectiveness is limited. According to Mondal et al. (2021), the minor impact of zeolite on crop yields suggests that its potential benefits (nutrient and water retention) may be more pronounced in less favourable soil conditions. This is particularly relevant given that most cultivated soils naturally exhibit low fertility due to limited nutrient availability. Low soil fertility contributes to reduced fertiliser use efficiency, particularly in the case of mineral fertilisers. Nutrient losses caused by various environmental factors result in inefficient nutrient uptake in light and sandy soils (Lipiński et al., 2020). Therefore, a key challenge in modern agriculture is the retention of nutrients in soil, which maximises their availability to plants while preventing groundwater contamination. In this context, zeolite applications in agriculture play a crucial role in soil fertility management (Babaousmail et al., 2022; Gondek, Mierzwa-Hersztek and Jarosz, 2023).

The study found that zeolite application resulted in only slightly increased nitrogen content in lettuce. Moreover, the different zeolite doses had minimal effect on nitrogen levels. Similar results were reported by Kavvadias et al. (2023), who found that zeolite application had little effect on nitrogen content in soil or lettuce leaves. This limited influence may be attributed to high cation exchange capacity, which can restrict the absorption of nitrates and ammonium nitrogen from the soil solution (Sangeetha and Baskar, 2016; Eroglu, Emekci and Athanassiou, 2017). However, Medoro et al. (2022) suggested that zeolite prolongs nitrogen retention in soil, facilitating plant uptake, which ultimately result in similar nitrogen absorption despite reduced input levels. Kavvadias et al. (2023) obtained similar results, showing that the addition of zeolite significantly increased nitrogen uptake regardless of soil type. The results are particularly relevant for light soils, considering the lower availability of potassium in these soils, especially at their low pH (Lipiński et al., 2020). It has been shown that the addition of 5% zeolite can significantly reduce potassium exchange in soil compared to lower doses (0 and 2%). This may reflect the strong

tendency of zeolite to adsorb K (Mondal *et al.*, 2021). In the present study, the effect of different zeolite doses on potassium content in lettuce leaves was minor, which may be due to its stabilising influence on bioavailable forms of potassium in the soil (Lipiński, 2019).

Phosphorus (P) deficiency in soils limits crop productivity in many regions of the world. It is estimated that P availability for plant roots is restricted in approximately two-thirds of cultivated soils worldwide, which poses a significant challenge to agricultural production (Lipiński, 2019). The use of phosphorus fertilisers in agriculture is a growing concern, as phosphate rock deposits, used in fertiliser production, are expected to be depleted within the 21st century. For this reason, optimising phosphorus fertilisation is one of the key challenges in modern agriculture, ensuring the conservation of this non-renewable resource (Tian et al., 2020). Furthermore, only 20% of the phosphorus applied as fertiliser is absorbed by plants and recovered in crops (Arrobas et al., 2021). Unfortunately, in the present study, different zeolite doses did not significantly affect phosphorus content in examined plants. Similar findings were obtained by Kavvadias et al. (2023), who reported that the addition of zeolite to the substrate did not influence P content in lettuce leaves. According to Hasbullah, Ahmed, and Majid (2020), zeolites effectively absorb phosphorus, limiting its availability to plants. A similar trend was reported by Assimakopoulou et al. (2020), where zeolites enhanced P nutritional status in plants compared to the control. Arrobas et al. (2021) suggested that natural zeolites are preferred in crop cultivation due to their high exchange capacity, which increases phosphorus uptake while reducing environmental contamination

In the present study, higher sodium levels were recorded in plants from zeolite-treated plots compared to the control. According to Kavvadias *et al.* (2023), zeolite's crystalline structure facilitates cation absorption, and natural zeolites contain high amounts of sodium, making it more available in the soil and thus more accessible to plants.

As previously demonstrated, zeolite slightly increased macronutrient concentrations in lettuce leaves (except for magnesium), while greater differences were observed for micronutrients. As the zeolite dose increased, Fe, Cu, and Zn content in lettuce leaves also increased. However, this contrasts with Kavvadias et al. (2023), who found that zeolite application had no significant effect on Zn content in plants, even though zeoliteenriched soil can reduce metal bioavailability. Similarly, studies by Kavvadias et al. (2023) and Arrobas et al. (2021) also reported a negligible effect of zeolite on other micronutrients, such as Cu. However, other researchers have noted that zeolite can reduce the uptake of Cu, Cr, Zn, and Cd in lettuce, spinach, and parsley (Cadar et al., 2022). Mondal et al. (2021) suggested that the limited effect of zeolite on micronutrient availability results from its ability to slow the release of nutrients from both organic and inorganic fertilisers, ensuring their availability over a longer period.

Natural and synthetic zeolites are recognised as soil amendments that reduce the bioavailability of heavy metals, particularly by significantly decreasing lead and cadmium solubility in contaminated soils (Hudcová *et al.*, 2021; Gondek, Mierzwa-Hersztek and Jarosz, 2023). Similarly, in the present study, lettuce leaves from plots treated with zeolite contained lower levels of cadmium and lead compared to the control. According to Kanagalabavi *et al.* (2024), zeolites are commonly

used for heavy metal sequestration (Cd, Pb, Cr, Zn, Cu, Fe, etc.) due to their high cation exchange capacity, which reduces metal mobility in the soil and, consequently, the uptake by plants. These properties of zeolite have also been explored by other researchers (Głąb, Gondek and Mierzwa-Hersztek, 2021).

To establish robust evidence on the effects of zeolite on both crop yield and quality, further studies should focus on different plant species and include long-term field trials with various soil types representative of the tested crops. Furthermore, the implementation of innovative crop management techniques is essential, not only to enhance soil water retention and nutrient availability, but also to reduce the leaching of nitrogen and phosphorus compounds into the environment (Lipiński *et al.*, 2020; Santos, Shinzato and Castanho, 2024; Senila and Cadar 2024).

# **CONCLUSIONS**

- 1. The addition of zeolite to the soil at concentrations of 1, 5 and 10% (10, 50, and 100  $g \cdot kg^{-1}$  of soil) significantly improved the emergence of lettuce seedlings. The effect of adding zeolite at 50  $g \cdot kg^{-1}$  of soil increased by 29.11% the emergence of lettuce seedlings, and there was no significant effect on each of 1 and 10  $g \cdot kg^{-1}$  of soil.
- As the zeolite content in the soil increased, both the yield of edible parts and the root biomass of lettuce also increased.
- Zeolite application had a beneficial effect, significantly reducing cadmium and lead concentrations in lettuce leaves.
- 4. The application of 10 g (1%) of zeolite per kilogram of soil led to a reduction in the nitrate content in lettuce leaves, while higher doses increased nitrate levels. However, all recorded values remained below the permissible limits.
- 5. Zeolite applied at lower doses (1 and 5%) had no significant effect on the content of flavonoids, anthocyanins, and polyphenols in the edible parts of lettuce, whereas a higher dose (10%) significantly reduced the levels of these compounds compared to the control (with the exception of anthocyanins).
- 6. The effect of zeolite on the content of major macronutrients in lettuce leaves was variable. No significant influence of zeolite on sodium content was observed. However, the application of zeolite at a 5% dose significantly affected the concentrations of other macronutrients, either increasing or decreasing their levels. A statistically confirmed increase in the content of micronutrients, such as iron, copper, boron, and zinc, was also demonstrated. In contrast, the concentrations of manganese and molybdenum decreased with the increasing zeolite doses in the soil.

# **CONFLICT OF INTERESTS**

All authors declare that they have no conflict of interests.

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