Albanian j. agric. sci. 2017;16 (2): 73-79 *Agricultural University of Tirana*

RESEARCH ARTICLE **(Open Access)**

The Influence of Natural Zeolite on Reygrass Growth in Heavy Metal Contaminated Soil

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Abstract

The use of natural zeolite can be a viable technique for the remediation of soils contaminated with heavy metals. This study aimed to evaluate the growth of reygrass (*Lolium multiflorum L.*) with different rate of zeolite in a soil contaminated with heavy metals generated by industrial activities. The rates of zeolite were as follows: 0, 1.25, 2.5, 5 and 10% (w/w), arranged in a completely randomized design with 4 replications. The plants were grown for 82 days in a greenhouse at the Agricultural University of Tirana. The results indicated that the addition of natural zeolite at all the application rates did not affect significantly ($p<0.05$) the germination of ryegrass seeds. However, the lowest germination performance (71.4%) was observed at the highest zeolite application rate (10%). This could be due to the effect of zeolite on electrical conductivity of the experimental soil. The plant height increased with increasing application rate of zeolite. The application of zeolite at 10% increased significantly (25.2%) the plant hight in the second cutting. Plant root and shoot dry weight was increased by increasing zeolite application, where the respective maximum increase was found at zeolite application rate of 10%. The root/shoot ratio of the plants was significantly correlated with the zeolite application rate, suggesting that the tested zeolite can functions as good plant growth regulator in heavy metal contaminated soil.

Keywords*:* chemical stabilization, heavy metal polluted soil, natural zeolite, reygrass

1. Introduction

Contamination of soils with heavy metals may pose risks and hazards to humans and the ecosystem [14]. The soils contaminated with heavy metals in Albania includes: industrial sites and *adjacent areas;* agricultural areas treated for a long time with pesticides, fertilizers and wastewaters [6], and soils naturally rich in heavy metals. According to Zdruli and Lushaj [15], about 16500 ha chemically polluted soils exist in the country. Only the industrial site of Rubik and its damp cover an area of 12.40 ha [10]. The soils close to this site are contaminated with cadmium, copper, nickel and zinc (F. Gjoka, personal communication), which are included in this study. The soils near industrial sites are increasingly used by local population for growing food plants.

Although some studies of these soils have been conducted [4; 5; 12], researches on the potentiality of soil amendments in the *immobilization* of heavy metals in heavy metal contaminated soils are missing. On the other hand, experimental data from the international literature on the use of natural zeolite for remediation purposes are also missing. Zeolite applications are based on their unique properties like adsorption, cation exchange, dehydration-rehydration, and catalytic properties [11]. It is found that zeolite application enhances plant yield (Castaldi *et al*., 2005, quoted by [9].

This research aimed to evaluate the growth of reygrass (*Lolium multiflorum L.*) with different rate of natural zeolite collected from Munella area (northern Albania) in a sandy soil contaminated with heavy metals generated by industrial activities.

2. Materials and methods

2.1 The study soil

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(Accepted for publication June 12, 2017)
***ISSN**: 2218-2020, © Agricultural University of Tirana*

The soil used in this study was Leptic Cambisols (Eutric) [3]. A composite soil sample (about 100 kg) was collected from the surface layer (0-25 cm) of contaminated area near a former copper smelter in Rubik, northern Albania (41°37' 2.984" N, 20°0'41.748" E). The air dried soil was crushed and passed through a 2-mm sieve prior to analysis and potting. The measurements of heavy metal concentrations in soil samples were made in the laboratories of the Department of Agrarian and Environmental Sciences, University of Udine, Italy (Table 1). The soil properties were described in detail by Gjoka et al. [5].

Table 1. Selected properties of the study soil

Soil parameter	Unit	Value
pH ($CaCl_2$)		6.71
Total-C	%	1.51
Clay	%	42.6
Cu	mg kg^{-1}	299
Ni	mg kg^{-1}	140
Zn	mg kg^{-1}	228
Cd	mg kg^{-1}	7.44

2.2 The zeolitic material

The zeolitic material as Stilbite-Stellerite used in this study was collected from the Munella deposits, northern Albania. Chemical composition of the zeolite was determined in the laboratories of the Soil Science and Soil Conservation Institute of Justus-Liebig University of Giessen, Germany (Table 2).

Table 2. Chemical composition of the zeolite used in the experiment

Property	Unit	Value
pH (H_2O, 1:5)		7.8
$CaCO_3$	%	<0.5
CEC_{pot}	meq/100g	6.72
EC (1:5)	µS/cm	91
K	mg kg^{-1}	3.91
Na	mg kg^{-1}	75.90
Ca	mg kg^{-1}	1196
Mg	mg kg^{-1}	33.60
Mn	mg kg^{-1}	ND
Fe	mg kg^{-1}	33.60

ND not detected

2.3 Experimental design

The pot experiment was carried out at the Agricultural University of Tirana, Albania, during April-June 2014. The experiment was laid out in a randomized block design with six treatments replicated four times. The sandy soil-zeolite composites were mixed at weight per weight percent (w/w %) ratios of: 100:0 (control); 98.75: 1.25, 97.5:2.5, 95:5, and 90:10. The plastic pots were filled with 1.5 kg of dry soil-zeolite mixture. The treated soils was incubated at about 70% of water-holding capacity (WHC) for 3 weeks, and pots then were planted with ryegrass (*Lolium multiflorium, L)* and kept in a greenhouse with a temperature of 25°C, being watered regularly, until the plants matured.

2.4 Observations and measurements of plants

During the experiment, phenological observations and biometric measurements of plants are performed, inluding plant germination, plant health (color), number and size of leaves, plant height, and dry weight of plants after each cutting. The plants were cut at 52 and 82 days after sowing.

2.5 Data analyses

The data were analyzed by ANOVA. The LSD test at p<0.05 was used to separate the differences between treatments and control.

3. Results and Discussion

3.1 Influence of natural zeolite on seed germination

The results of ANOVA indicated that the effect of zeolite application on germination percentage was insignificant (p<0.05) (Table 3). However, germination percentage decreased with increasing rate of zeolite (Fig. 1). This phenomenon can be explained by increases in electrical conductivity of the experimental soil with increasing application rate of zeolite. Zhang et al. [16] reported that salinity negatively affected seed germination.

Table 3. Results of ANOVA for seed germination

Source of Variation	SS	df	MS	F	F crit	p-value
Replication	1914,65	4	478,66	1,84	3,05	0,17
Zeolite rate	3891,71	15	259,44			
Total	5806,36	19				

In contrast, Ajirloo et al [1] *found* that increasing the zeolite (Clinoptilolite with high CEC) application rate significantly decreased electrical conductivity of soil. The zeolite (*Stilbite-Stellerite)* used in the study had low CEC (6.72 meq/100g). Application of zeolite at the rate 10% (w/w) led to greatest decrease in the percentage of seed germination (79%). In zeolite treated soils, maximum percentage of germination was obtained by addition of zeolite at the rate 1.25%. Khan et al. [7] indicated that zeolite application at the rate of 1 and 2% improves plant seed germination.

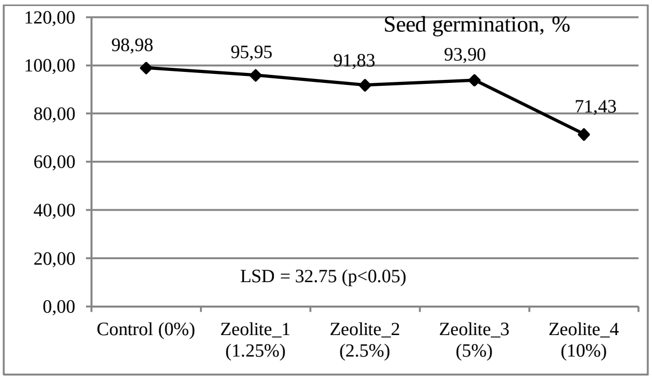

Figure *1*. *Effect* of zeolite rate on seed *germination*

3.2 Influence of natural zeolite on plant hight

The ANOVA indicated that the effect of zeolite application on plant height was significant (p<0.05) in the second cutting, and insignificant in the first cutting (Tables 4-5). This *may* be *due* to *nutrient depletion* and loss of *soil fertility* after the first cutting, especially in the control treatment. Under these conditions, the effect of zeolite on plant growth becomes much more apparent.

Table 4. Results of ANOVA for plant hight (cutting I)

Source of Variation	SS	df	MS	F	F crit	p-value
Replication	2,87358	4	0,718395	0,405591	3,055568	0,80175
Zeolite rate	26,56848	15	1,771232			
Total	29,44206	19				

Table 5. Results of ANOVA for plant hight (cutting II)

Source of Variation	SS	df	MS	F	F crit	p-value
Replication	15,83817	4	3,959543	3,725547	3,055568	0,026821
Zeolite rate	15,94213	15	1,062808			
Total	31,7803	19				

As compared to control, the increase in plant height in the first cutting varied from 3.07% to 7.03% at the application rate of 1.25% and 10%, respectively (Fig 2). Similar results have been reported by [13]. While in the second cutting, the increase in plant height varied from 5.51% to 25.19% at the application rate of 5% and 10%, respectively (Fig. 2). Application of zeolite at the rate 10% (w/w) resulted in a significant increase in the plant height compared to control.

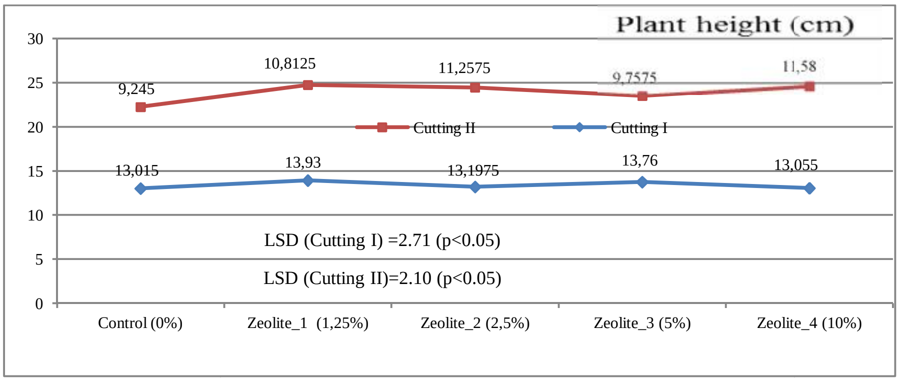

Figure 2. Effect of zeolite rate on plant height

3.3 *Influence of zeolite on dry weights of shoots and roots*

Unlike plant height, ANOVA indicated significant effect ($p<0.05$) of zeolite application on shoot dry weight of ryegrass in two cuttings (Tables 6-7). However, a greater effect was produced by zeolite in the second cutting (see F-tests). It seems that zeolite application improves plant growth and biomas production.

Table 6. Results of ANOVA for shoot dry weight (cutting I)

Source of Variation	SS	df	MS	F	P-value	F crit
Replication	0,07283	4	0,018208	3,308449	0,03932	3,055568
Zeolite rate	0,08255	15	0,005503			
Total	0,15538	19				

Table 7. Results of ANOVA for shoot dry weight (cutting II)

Source of Variation	SS	df	MS	F	P-value	F crit
Replication	0,39392	4	0,09848	8,019544	0,00115	3,055568
Zeolite rate	0,1842	15	0,01228			
Total	0,57812	19				

As shown in Figure 3, the application of zeolite at the rate 10% resulted in a statistically significant increase in the shoot dry weight in the first cutting compared only to zeolite application at the rate 2.5%. It seems to be related to other factors than zeolite application. While the application of zeolite at the rates 10% and 1.25% resulted in a significant increase of shoot dry weight in the second cutting compared to control (Fig. 3). These data suggest that application of zeolite in the studied soil improves the efficiency of mineral fertilizers.

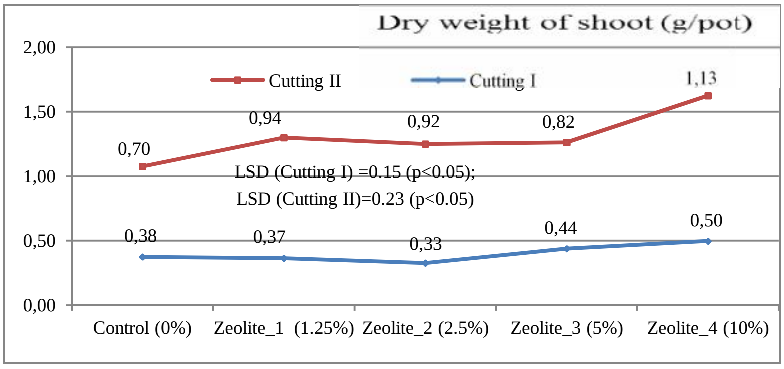

Figure 3. Effect of zeolite rate on shoot dry weight

The differences in the shoot dry weight between the treatments with zeolite can be explained by changes in *soil* nutrient status. According to Ming & Allen 2001, quoted by [8], the zeolite added to soil adsorbs the NH_4^+ ions during the incubation period. In this experiment, the soil was incubated for 10 days at *70* % of the maximum water holding capacity. In the wet soil, the NH_4^+ ions transferred from zeolite structure to soil solution as a result of ion-exchange by K^+.

The ANOVA results indicated that the effect of zeolite on root dry weight was insignificant (p<0.05), compared to control (Table 8). But, root dry weight increased with increasing zeolite rate from 1.25% to 10% (Fig. 4). The maximum value of root dry weight was obtained in treatment with 10% zeolite.

Table 8. Results of ANOVA for root dry weight

Source of Variation	SS	df	MS	F	P-value	F crit
Replication	6,553847	4	1,638462	1,371738	0,290391	3,055568
Zeolite rate	17,91663	15	1,194442			
Total	24,47048	19				

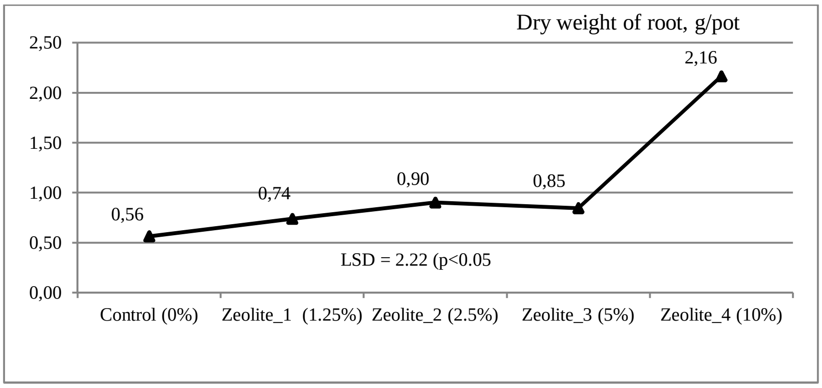

Figure 4. Effect of zeolite rate on root dry weight

Results of correlation analysis (Table 9, Fig. 5) indicated that the dry weight of shoot (cutting I plus cutting II) was positively and significantly correlated with dry weight of roots (r = 0.952, p <0.05;). This is fully justified from the point of view of the plant biology.

Table 9. Correlation *between* dry weights of shoot and root

	Dry weight of shoot	Dry weight of root
Dry weight of shoot	1	0.952*
Dry weight of root	0.952*	1

Correlation is significant at the 0.05 level (2-tailed)

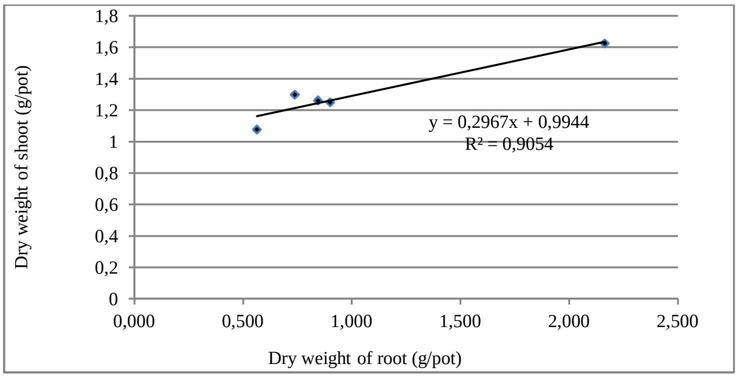

Figure 5. Relationship between shoot and root dry weight

In order to evaluate the relative growth of ryegrass, correlation coefficient between total dry weight and root dry weight was calculated (Table 10 and Figure 6). The r value (0.997**, p<0.001) suggests close relationship between root growth and the whole plant growth [2].

Table 10. Correlation *between* total dry weight and root dry weight

	Dry weight of shoot	Dry weight of root
Dry weight of shoot	1	0.997**
Dry weight of root	0.997**	1

Correlation is significant at the 0.01 level (2-tailed)

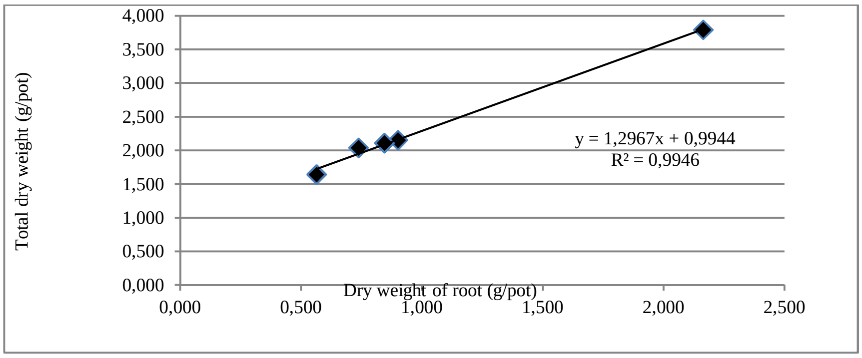

Figure 6. Relationship between total and root dry weight

Table 11 and Fig. 7 indicated that zeolite rate (%, w/w) was positively and significantly correlated with root/shoot ratio ($r = 0.942$, $p < 0.05$).

Table 11. Correlation *between* dry weights of shoot and root

	Zeolite rate	Root/shoot ratio
Zeolite rate	1	0.952*
Root/shoot ratio	0.952*	1

Correlation is significant at the 0.05 level (2-tailed)

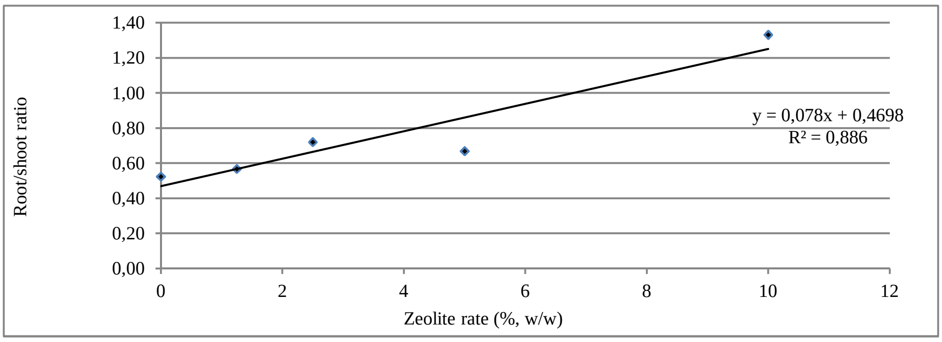

Figure 7. Relationship between zeolite rate and root/shoot ratio

4.Conclusions

Application of zeolite to a heavy metal contaminated soil had significant effect on the plant growth parameters like height, shoot dry weight and root/shoot ratio. The highest plant height, shoot and root dry weight and root/shoot ratio for reygrass in this study was observed when 10% zeolite was applied. The root/shoot ratio of the plants was significantly correlated with the zeolite application rate. Thus, it can be concluded that the tested zeolite can functions as good plant growth regulator in metal contaminated soil.

5. Acknowledgments

The research was supported by the Science and Technology (S&T) Cooperation Programme between the Republic of Italy and the Republic of Albania (2012 – 2014). We also thank the staff of the Soil Science and Soil Conservation Institute of Justus-Liebig University of Giessen (Germany) for its technical assistance in analyses of zeolite samples.

6. References

1. Ajirloo, G.S, Jadid, A.P., Nasirtabrizi, M.H., Mansoor, S.R: **Effect of zeolite application on soil purification and some chemical properties of soil (case study)**. Tech J Engin & App Sci. 2013, **3**(11): 970-974.
2. Fageria N.K., Moreira A: **The role of mineral nutrition on root growth of crop plants**. **In: Advances in Agronomy** Vo. 110, ed. D.L. D.L., Sparks (San Diego: Elsevier Academic Press), 2011: 268.
3. FAO: **World reference base for soil resources**. World Soil Resources Reports 103; 2006.
4. Gjoka, F., Felix-Henningsen, P., Wagener Hans-R: **Heavy metals in soils of vicinity of industrial Site of Laç, Albania**. Balkan Ecology 2002, **5**(3):301-306.
5. Gjoka, F., Salillari, I., Cenameri, M., Llaveshi, P: **Status of heavy metals of soils near the industrial complexes in Laç, Rubik and Burrel.** Albanian Journal of Agricultural Sciences 2007, **9**(6): 35-41.
6. Kasa, E., Contin, M., Gjoka, F: **Accumulation of Heavy Metals in Vegetables from Agricultural Soils**. **Albanian Journal of Agricultural Sciences 2015, 14(2): 169-175.**
7. Khan H., Khan A.Z., Khan R., Matsue N., Henmi T: **Influence of zeolite application on germination and seed quality of soybean grown on allophanic soil.** Research Journal of Seed Science 2009, **2**(1): 1-80.
8. Leggo, P.J: **Enhancing the Growth of Plants on Coal Waste Using a Biological Fertilizer**. International Journal of Environment and Resource (IJER) 2013, 2(3): 59-66.
9. Mahmoodabadi M.R., Ronaghi A., Khayyat M., and Hadarbadi Gh: **Effects of zeolite and cadmium on growth and chemical composition of soybean (Glycine max L.).** Tropical and Subtropical Agroecosystems, **10**(2009): 515 - 521
10. Mansaku (Meksi), M., Šamánek, J., Jopková M: **Pops hotspot infolist**: **Former copper plant, Rubik, Albania** 2009. http://english.arnika.org/files/documents/Rubik_hotspot_report.pdf
11. Mumpton F.A: **La roca magica: uses of natural zeolites in agriculture and industry**. Proc Natl Acad Sci U S A 1999 Mar, **96**(7): 3463–3470.
12. Sallaku, F., Fortuzi , Sh., Tota, O., Huqi, B., Chachalis, D., Darawsheh, M: **Heavy metal soil contamination around the metallurgical plant of Elbasani in Albania**. Journal: Food, Agriculture and Environment (JFAE) 2009, **7**(3&4): 878-881.
13. Türk, M., Bayram, G., Budakli, E., Celik**, N: A study on effects of different mixtures of zeolite with soil rates on some yield parameters of alfalfa (Medicago sativa L.), Journal of Agronomy 2006, 5(1):118-126.**
14. Wuana, R. A. and Okieimen, F. E: **Heavy Metals in Contaminated Soils: A Review of Sources, Chemistry, Risks and Best Available Strategies for Remediation**. International Scholarly Research Network ISRN Ecology 2011, Volume 2011, Article ID 402647, 20 pages doi:10.5402/2011/402647
15. Zdruli P., Lushaj S: **The status of soil survey in Albania and some of its major environmental findings. In: Zdruli P. (ed.) , Steduto P. (ed.) , Lacirignola C. (ed.) , Montanarella L. (ed.).** Soil resources of Southern and Eastern Mediterranean countries. Options Méditerranéennes: Série B. Etudes et Recherches, 2001, n. 34: 69- 89.
16. Zhang, H., Irving, L.J., McGill, C., Matthew, C., Zhou, D., Kemp, P: **The effects of salinity and osmotic stress on barley germination rate: sodium as an osmotic regulator**. Annals of Botany 2010, 106: 1027–1035.