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Dedicated to Professor Emil Cordoș
on the occasion of his 80th anniversary

THE INFLUENCE OF POLLUTED SOIL AERATION IN THE PROCESS OF IN SITU BIOLEACHING

IOANA MONICA SUR[a,*], VALER MICLE[a], TIMEA GABOR[a]

ABSTRACT. The influence of aeration of soils polluted with heavy metals by using *Thiobacillus*-type microorganisms was studied using soil samples contaminated with heavy metals (Cu 4074 - 7550 $mgkg^{-1}$, Zn 5870 - 9310 $mgkg^{-1}$, Cd 36-50 $mgkg^{-1}$, Pb 15000 - 42890 $mgkg^{-1}$), from Romplumb, Baia Mare. The variation of the metal concentration extracted by bioleaching and aerated bioleaching was monitored for 16 weeks. The soil samples treated by bioleaching (Cu: 9 - 53%; Zn: 9 - 62%; Cd: 9 - 24%. Pb: 31 - 71%) have obtained a lower efficiency than the soil samples treated by aerated bioleaching (Cu: 34 - 70%; Zn: 36 - 76%; Cd: 17 - 38%. Pb: 44 - 78%), but there are percentage differences between the two processes (Cu: 17 - 27%; Zn: 14 - 27%; Cd: 8 - 14%, Pb: 7 - 13%).

Keyword: *bioleaching, contaminated soils, heavy metals, in situ, microorganisms.*

INTRODUCTION

Soil pollution with heavy metals in Baia Mare is recognized today as a significant problem and is a major risk to human health and the environment [1 - 3]. High concentrations of heavy metals in soils are related to anthropogenic pollution sources, mining and metallurgy [4].

Heavy metals pollution has a cumulative and residual character, which means that pollutants accumulate slowly, being the result of a permanent and long-term exposure of the soil to the action of these pollutants [5, 6].

[a] *Technical University of Cluj-Napoca, Faculty of Materials and Environmental Engineering, Department of Environmental Engineering and Sustainable Development Entrepreneurship, 103-105 Muncii Ave, Cluj-Napoca, Romania*
* *Corresponding author: ioanamonica_berar@yahoo.com*

Bioleaching, or bacterial leaching, consists in the extraction by solubilization of the metallic elements from contaminated soil using bacteria. This method does not destroy (eliminate) the pollutants, but it favors their segregation from the contaminated environment, the microorganisms having the property to oxidize the metals, transforming them into a more soluble form [7 – 8].

Thiobacillus ferrooxidans was successfully adapted to leaching of both Cu, Ni, Fe, Zn, Cd [9 - 12].

There are several studies in the literature related to bioleaching of metals and metalloids using different type of microorganisms [13 - 23]. *Thiobacillus ferroxidans* give high yields in case of bioleaching of Cu, Cd and Zn [13, 14, 16, 18, 20 - 23], while *Thiobacillus thiooxidans* was proven to be efficient in the extraction of Cd, Cr; Cu, Mn, Ni, Pb, Zn [15, 17, 19]. However, bioleaching is still under study in order to determine the influence of the different parameters which can result in a higher yield and efficiency.

The preliminary experimental laboratory results have shown high extraction yields: Fe: 100%; Cr: 11- 25%; Cu: 40 - 100%; Zn: 55 - 94%; Cd: 17 - 60% și Mn: 32 - 66% [18].

The present researches and studies are focused on in situ bioleaching; thus, it was considered a necessity to develop a research that approaches to the method of treating the soil polluted with heavy metals by in situ bioleaching applying also one of the main parameters (soil aeration) which leads to high treatment efficiency.

The main advantage of in situ bioleaching is that the entire process happens without soil excavation, so the transportation costs are eliminated and the transport does not pollute the environment. In this way, capital and energy costs are low [24].

The purpose of this paper is to observe the influence of aeration in the extraction of heavy metals from polluted soils by in situ bioleaching, using *Thiobacillus ferrooxidans* type of microorganisms in 9K medium.

The paper tracks how aeration influences the in situ bioleaching process, applied to extract heavy metals from soil used in the experimental research. This is evidenced by variation in metal concentration depending on time and depth.

RESULTS AND DISCUSSION

Variation of extracted metal concentration

Copper is showing a very good extraction after only 4 weeks of experiment in the depth 0 – 10 cm. In the case of the sample extracted from 10 – 20 cm depth, it can be seen a decrease of Cu achieving maximum extraction after 8 weeks (Fig. 1).

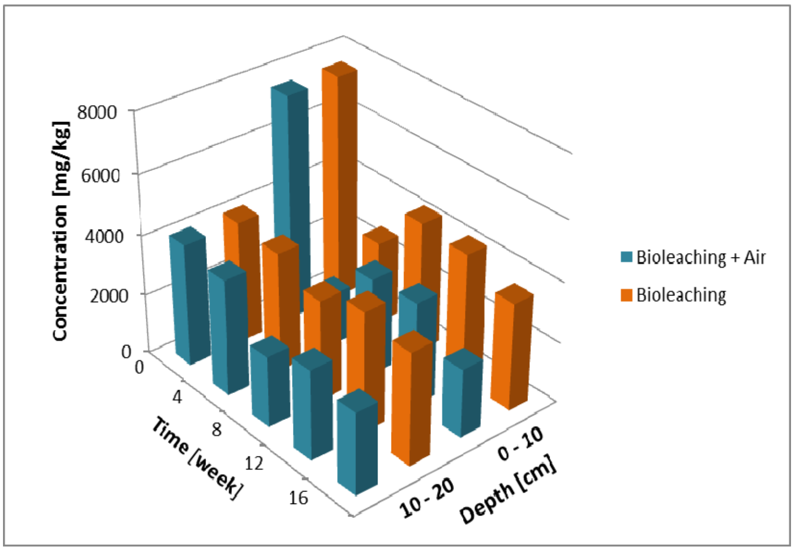

Figure 1. Variation in time of copper concentration in soil

We have seen maximum Zn extraction (Fig. 2) after 8 weeks of experiment, on both soil samples, regardless of the depth of sampling and the decontamination method applied (bioleaching or bioleaching + aeration).

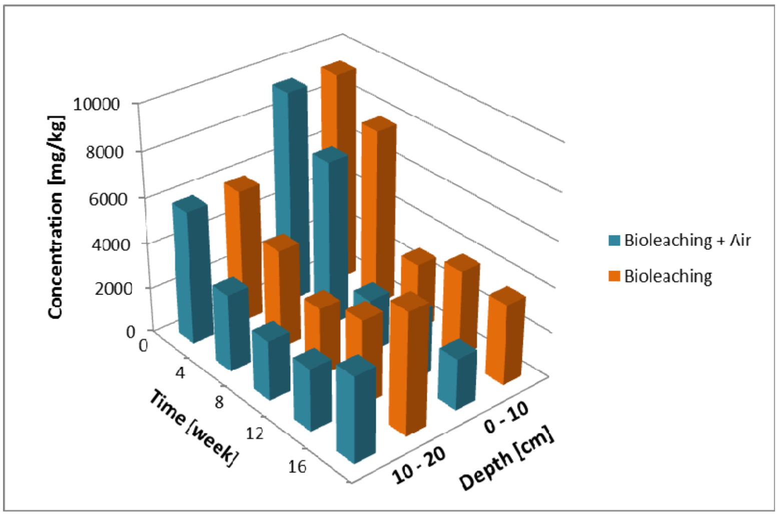

Figure 2. Variation in time of zinc concentration in soil

Cadmium and lead both have a good extraction after 8 weeks of experiment, regardless of the sampling depth (Fig. 3).

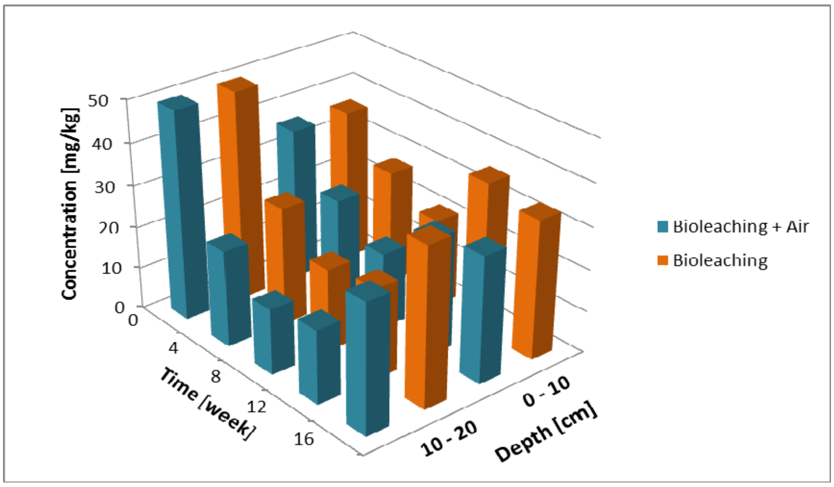

Figure 3. Variation in time of cadmium concentration in soil

The soil samples submitted to bioleaching + aeration show smaller concentration of lead than the soil samples submitted only to bioleaching during the entire period of experiment, regardless of the depth of sampling (Fig. 4).

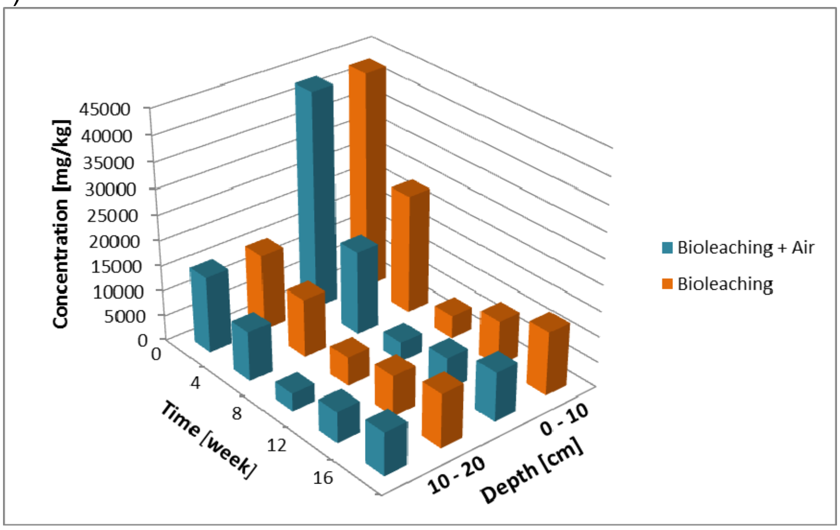

Figure 4. Variation in time of lead concentration in soil

The increase of Cu, Zn, Cd, and Pb metal concentration after 12 weeks is due to the metabolic activity of the microorganisms.

Both experiments have the same extraction trend, with a better extraction for the aerated soil samples.

Efficiency of extraction process

Evaluation of the extraction process efficiency for the metals in the soil after 16 weeks was carried out by determining the final yield of each element, by the following formula [25]:

$$\eta = \frac{C_i - C_f}{C_i} 100 \; [\%] \tag{1}$$

where: η final yield, in %;

C_i – initial metal concentration in soil sample, in mgkg^{-1};

C_f – metal concentration in soil sample at final treatment time, in mgkg^{-1}.

Analyzing figure 5 can be seen that Cu, Zn and Pb are showing a very good extraction (70%), Cd having the lowest leaching (17%).

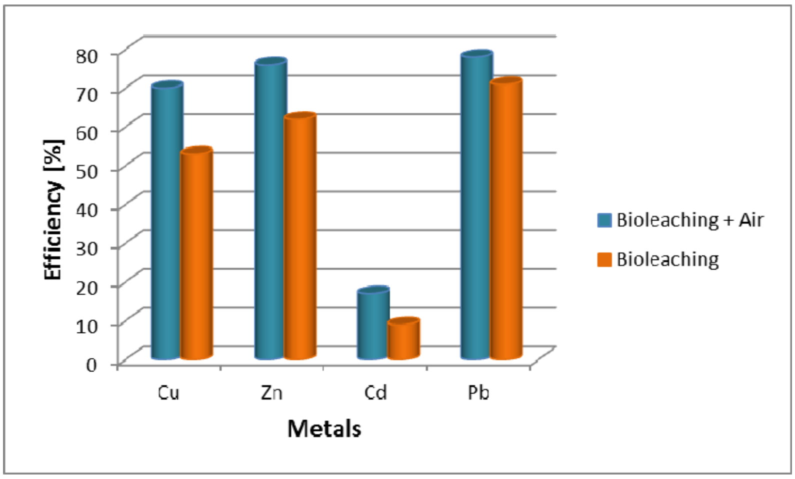

Figure 5. Efficiency of metals extraction from depth 0 – 10 cm

In figure 6 is showing that lower yields have been obtained (34 - 44%) compared with the surface sample. This is due to the fact that the metals from the surface sample were washed with leached solution in the moment when were added on top of the soil samples.

Comparing the two methods, the extraction efficiency of metals is much higher if aeration is introduced in the process (Cu: 17 - 27%; Zn: 14 - 27%; Cd: 8 - 14%. Pb: 7 - 13%).

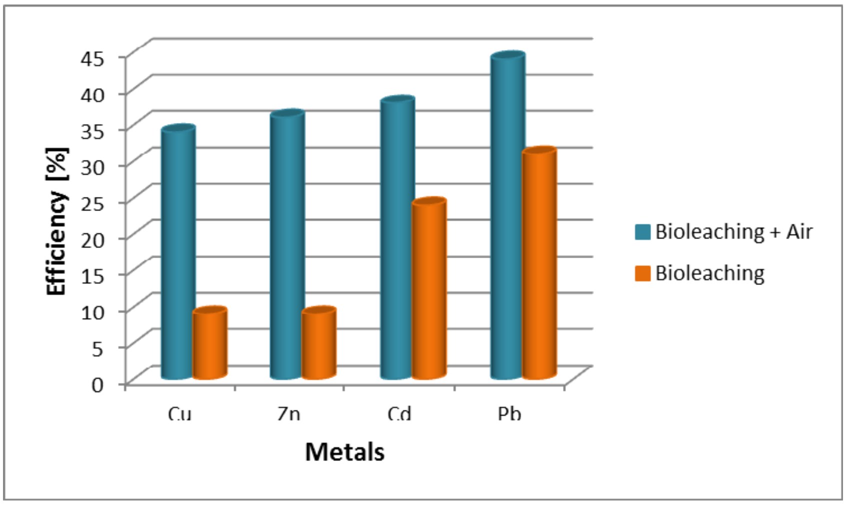

Figure 6. Efficiency of metals extraction from depth 10 – 20 cm

The Cu (34 - 70%) extraction yields are similar with the ones presented by Couillard and colleagues [21], but lower when compared with other results from the literature [14, 19, 22, 23].

Zinc (36 - 76%) has a lower extraction yield compared with the studies of Blais and colleagues (88 - 97%) [19].

The extraction yield for Cd (17 - 38%) is considerably lower than the ones from other experiments (80%) [19, 21, 23].

Lead has an extraction yield better than the one obtained in the experimental researches of Blais and colleagues (10 - 54%) [19].

CONCLUSIONS

The variation in the extracted concentration of heavy metal from soil samples shows a linear decrease of metals in soil, a good extraction being obtained after eight weeks of leaching.

The bioleaching solution used has a better efficiency for the extraction of metals such as Pb, Zn, Cu, but lower efficiency in the case of Cd extraction.

The extraction efficiency is higher for surface soil layer (0 - 10 cm depth) in case of all metals which have migrated underground due to the bioleaching solution that was added weekly over the soil samples.

Higher concentrations of metals compared to the samples subjected only to bioleaching were extracted from the samples subjected to both bioleaching and aeration.

The quantity of metal in the soil shows the same variables, whichever method is applicable (bioleaching or bioleaching + aeration) and regardless of the depth which was taken from the ground.

The results indicated that the bioleaching of metals is achieved by the growth of *Thiobacillus* bacteria type. After 16 weeks of treatment of bioleaching + aeration, heavy metals were extracted from soil metals, as follows: Cu: 34 - 70%; Zn: 36 - 76%; Cd: 17 - 38%; Pb: 44 - 78%.

The extraction efficiency of metals is much higher if aeration is introduced in the process (Cu: 17 - 27%; Zn: 14 - 27%; Cd: 8 - 14%. Pb: 7 - 13%).

The experiments carried out have led to the conclusion that soils can be decontaminated using in situ bioleaching and if aeration is used, higher yields are obtained.

EXPERIMENTAL SECTION

Sampling

Soil samples in natural state used for this lab experiment were collected from Romplumb, Baia Mare. The sampling point is located in the vicinity of the technologic gas chimney area and laboratories. From the research area were taken two soil samples in natural condition, as follows: **sample 1**, depth: 0 – 10 cm and **sample 2**, depth 10 – 20 cm. The samples in natural or undisturbed state were collected in a special container that keeps the soil natural settlement mode [26 –28].

Culture media

Remediation of contaminated soils was performed using *Thiobacillus ferrooxidans* type microorganisms (140 x 10^6 cells/mL), selected from the soil sampling area (Baia Mare). These microorganisms were grown on a 9K type nutrient medium with the following solutions: $(NH_4)_2SO_4$ - 3 g; KCl - 0.1 g; K_2HPO_4 - 0.5 g; $MgSO_4 \cdot 7H_2O$ - 0.5 g; $Ca(NO_3)_2 \cdot 4H_2O$ - 0.01 g; $FeSO_4 \cdot 7H_2O$ - 44.2 g is dissolved in distilled water (1 L), yielding a solution with pH = 2.5 [29].

Experimental

The soil samples taken in their natural state were transferred to glass containers (Lxlxh=150mmx150mmx300mm) and placed on a drainage layer of 30 - 45 mm gravel sort, depending on the depth of the sample in order to reconstruct the profile of the soil. Bioleaching solution – 9K medium, was added over the prepared soil samples, the soil samples being dipped in the bioleaching solution, to which was added on a weekly basis 100 ml of solution.

The process of cleaning the polluted soils with heavy metals was done by: bioleaching and bioleaching + aeration, the two processes being performed independently. Both experiments were done in the same conditions: pH=5, constant temperature (26 ± 2°C) and soil dipped in bioleaching solution. In the case of in situ bioleaching with aeration was introduced a perforated tube which was connected to a compressor, achieving 8 bar aeration pressure for 8 hours/a day on the entire duration of the experiment (16 weeks). Concentration from week 0 represents the quantity of pollutant present in the soil before beginning the remediation process.

After the start of the experiment, soil samples (10 g) were collected once per month for four months (16 weeks) in order to establish the speed of metal extraction from soil in the process of in situ leaching and to observe the influence of soil aeration in the process of bioleaching.

Metal concentration in soil samples were analyzed by inductive coupled plasma atomic spectrometry - ICP-AES (Optima 5300DV, Perkin Elmer) of the Research Institute for Analytical Instrumentation (ICIA-Cluj-Napoca).

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REFERENCES

1. ***, Annual report on the state of the environment in Romania in 2007, the National Agency for Environmental Protection, Bucharest, **2008**.
2. M. Coman, V. Oros, B. Fălăuş, R. Pop, *ProEnvironment/ProMediu,* **2010**, 3, 155–158.
3. C. S. Cociorhan, V. Micle, V. Oros, I. M. Berar-Sur*, ProEnvironment/ProMediu*, **2011**, 4, 179–186.

4. F. Damian, G. Damian, R. Lăcătuşu, G. Macovei, G. Iepure , I. Năprădean, R. Chira, L. Kollar, L. Raţă, D. C. Zaharia, *Carpth. J. of Earth and Environmental Sciences*, **2008**, *3(1),* 85–98.
5. L. Miloiu, "Actual status of the environment in the Baia Mare and Copalnic basin"–PhD Thesis (in Romanian), Oradea, **2008**.
6. I. M. Sur, V. Micle, G. Plopeanu, *Ecoterra – Journal of Environmental Research and Protection,* **2012**, *31*, 105–109.
7. I. M. Sur, *Acta Technica Napocensis,* **2015***, 4(4), 61–67.*
8. ***, Feasibility study – Eco Industrial Parc SEPA Baia-Mare (in Romanian), Baia Mare, **2012**.
9. L. J. Mason, N.M. Rice, *Minerals Engineering*, **2002**, *15*, 795–808.
10. S. Kiss, D. Paşca, M. Drăgan-Bularda, "Enzymology of disturbed solis Developments in Soil Science", Editura Elsevier, **1998.**
11. T. Volke Sepúlveda, J.A. Velasco Trejo, D.A. de La Rosa Pérez, "*Suelos contaminados por metales y metaloides: muestreo y alternatives para su remediación*", Méxic, **2005.**
12. S. N. Groudev, I. I. Spasova, P. S Georgiev, *International Journal of Mineral Processing*, **2001**, *61(1*), 301–308.
13. X. Xuehui, Y. Xuewu, N. Liu, C. Xiaoguang, A. Awad, L. Jianshe, *BioMed Research International*, **2013**, 1–10.
14. D. B. Nakade, *International Journal of Advance Research in Engineering and Technology* (IJARET), **2014**, *5(1),* 52–58.
15. R. Naresh Kumar, R. Nagendran*, J Hazard Mater*, **2009**, *169*, 1119–1126.
16. H.-L. Liu, B.-Y. Chen, Y.-W Lan, Y.-C. Cheng, *Appl Microbiol Biotechnol*., **2003**, *62*, 414–420.
17. B.-Y. Chena, H.-L. Liu, Y.-W. Chenb, Y.-C. Chengb, *Process Biochemistry*, **2004**, *39*, 735–745.
18. I.M. Berar-Sur, V. Micle, S. Avram, M. Şenilă, V. Oros, *Environmental Engineering and Management Journal*, **2012**, *11(8*), 1389–1393.
19. J. F. Blais, R. D. Tyagi, J. C. Auclair, *Water Research*, **1993**, *27*, 101–110.
20. A. T. Lombardi, G. Jr. Oswaldo, *Water Research*, **2002**, *36*, 3193–3202
21. D. Couillard, M. Chartier, G. Mercier, *Journal of Water Science*, **1994**, *7,* 251–268.
22. R. C. Ivănuş, G. Bică, *Studia Universitatis "Vasile Goldiş" Series: Economic Sciences*, **2007***, 12,* 109–117.
23. R. C. Ivănuş, "Bioleaching Solubilization" (in Romanian), Universitaria Press, Craiova, Romania, **2005**.
24. Abhilash, B. D. Pandey, K. A. Natarajan, "Microbiology for Minerals, Metals, Materials and the Environment", CRC Press Taylor&Francis Group, **2015.**
25. A. Măicăneanu, H. Bedelean, M. Stanca, "Natural zeolites–Characteristics and applications in environmental protection" (in Romanian), Presa Universitară Clujeană, **2008**.
26. ***, STAS 7184/1-84, Soils. Sampling for soil and agrochemical studies (in Romanian).
27. ***, Order no. 148 of 09/21/1997, approving the procedure of environmental audit, Official Gazette no. 303 of 06.11.1997 (in Romanian).

28.***, Order no. 756 of 11.03.1997 approving the regulations on environmental pollution assessment, Official Gazette no. 303 of 11/06/1997 (in Romanian).
29.M. P. Silverman, D. G. Lundgren, *Journal of Bacteriology*, **1959**, *77*, 642–647.