Barranquilla - Colombia - June 21st and 22nd 2018

7th Academic INTERNATIONAL WORKSHOP ADVANCES IN CLEANER PRODUCTION

"CLEANER PRODUCTION FOR ACHIEVING SUSTAINABLE DEVELOPMENT GOALS"

Recovery of Heavy Metals from Waste Printed Circuit Boards Through Microbiological Leaching, Using Consortia of Acidophilic Chemolithotrophic Bacteria

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Abstract

An alternative to reduce environmental impact and cost in the extraction of metals from electronic waste is the use of bacterial leaching processes. In this work, the recovery of heavy metals from wasted printed circuits boards (WPCBs) of desktop computers through bacterial leaching processes has been investigated. Consortia of chemolithotrophic acidophilic bacteria were obtained from acid water and rocks from a local mining action, and from microorganisms isolated from WPCBs. We used X-ray fluorescence spectroscopy to quantify the amount of metals present in WPCBs before, during and after exposure with the isolated bacterial study consortia. Growth conditions of the microorganisms were studied, metal leaching rate present in the WPCBs by these consortia was determined under different conditions of pH, temperature and agitation in several bioassays. This study demonstrated the bioleaching of toxic metals such as lead, nickel and chromium, as well as other metals such as iron, calcium, zinc, manganese, copper, osmium, tantalum, platinum, and gold.

Keywords: *Bioleaching, heavy metals, chemolithotrophic bacteria, printed circuit boards (PCB), adaptation*

Introduction

Waste from printed circuit board (WPCBs) of discarded computers represents an important resource of metals pollutants in the environment (Chen et al., 2015). They constitute 3 to 5% by weight of the total waste electrical and electronic equipment (WEEE) (Montero, 2012); In addition, within this electronic component is the highest concentration of precious, heavy and strategic metals such as copper, aluminum, nickel, iron, tin, lead and precious metals such as silver and gold (Willner, 2013). However, WPCBs also contain a large number of hazardous substances such as some heavy metals and brominated retardants (Zhou, X. et al., 2013). Therefore, the recycling and decontamination of WPCBs are necessary for the protection of the environment (Chen et al., 2015).

The most used technologies for the treatment of WPCBs include pyrometallurgical and hydrometallurgical processes (Oliveros, 2011). However, the process of recycling WPCBs using these techniques can hardly satisfy economic requirements, not even the simple technology of production and management of the green processes that have been developed in the last years (Yagnentkovsky,

2011). Bioleaching is a promising technology which utilizes microorganisms to recover metals from low grade ores and e-waste (Chen, S et al. 2015). Benefiting from lower operational cost and energy requirements in metal recovery, bioleaching has drawn more and more attention. Chemolithoautotrophs bacteria, such as *Acidithiobacillus ferrooxidans*, *Leptospirillum ferrooxidans*, *Acidithiobacillus thiooxidans*, etc, are commonly used for metal recovery from e-waste (Willner, 2013; Rodríguez, E., 1994). Those bacteria which use CO_2 as carbon source and inorganic compounds (Fe^{2+} and reduced S) as energy source, utilize their ability to facilitate metal dissolution through a series of biooxidants and bioleaching reactions.

Previous studies (Wu, W. et al. 2018; Bryan, C.G., et al., 2015; Chen, 2015; Willner, 2013; Zhou, X., et al., 2013) indicate that the process of metals bioleaching from electronic waste is viable, although it is a complex process, determined by many factors, including: type of microorganisms, concentration of Fe^{2+} in the system, qualitative and quantitative composition of waste, toxicity of ingredients and fineness of the material. Temperature, pH and time also play a significant role in the reaction. In addition, it can be assumed that the mechanism of bioleaching of metals from electronic scrap involving bacteria is the same, as in the case of metal sulfides leaching, direct and indirect leaching (Choi, Ms., et al., 2005).

The purpose of the present study was to demonstrate that it is feasible to achieve the bioleaching of heavy metals (Pb, Ni, Cr) from WPCBs using a Chemolithoautotrophs microbial consortium extracted at mining sites. This process was carried out controlling parameters of temperature, initial pH and speed of agitation as a function of time and the growth conditions of microorganisms were studied under these circumstances. In addition, different factors that affect the leaching process of heavy metals and the inhibition of microbial growth were explored.

2. Materials and methods

2.1. Preparation of integrated circuits samples

The WPCBs used throughout this study belonged to Board Intel DDR3. Only pieces of this specification were taken in order to maintain the proportions of the metals present in the pieces. The total sample weighed 2200 g. For experimental use of microbial leach, the pieces were ground to a size of 0.5 cm after manually removing the main electronic components (for example: capacitors and resistors). Once the pieces were crushed, they were introduced into aqua regia, forming highly concentrated liquor with the metals of the solution pieces from which three aliquots were taken, which were tested by total X-ray fluorescence spectrometry (TXRF) using an S2 PICOFOX (Bruker), where concentrations of metallic elements present in the WPCB sample were measured. Based in the obtained findings, new pieces of size 0.5 cm were isolated and these would be later incorporated in solid state to the different microbial consortia with the intent of identifying the lixiviated metals in this liquor and compare them with the metals detected initially in the aqua regia.

2.2. Collection of samples

The source of microorganisms used in this study consisted essentially of three microorganism consortia from: **A)** acid waters; **B)** rocks; and **C)** microorganisms housed on the surface of WPCBs. Samples **A** and **B** were collected in local mining zones.

2.3. Obtaining consortia, adaptation and culture conditions

Culture adaptation consisted of two phases: In phase 1, microorganisms present in WPCBs were isolated from a portion of these circuits equivalent to 12 g, inoculated into 90 ml of nutrient broth (DIBICO), and incubated in the dark at 35 °C for 48 hours for potentiating the growth of microorganisms that might be present in the sample and which required moisture and availability of essential nutrients. Afterwards, they were re-grown in liquid medium, inoculating 10 ml of the stock culture in 125 ml Erlenmeyer flasks with 90 ml of nutrient broth (DIBICO), at pH 2 and 1.5, restocking

the new cultures every 48 hours during two weeks. All culture broths were prepared and sterilized in autoclave according to standard procedures (Atlas, R. 2004).

Samples of water and rock collected from the mine were subjected to a first adaptation in Silverman and Lundgren's 9k medium, which was prepared from two solutions (Merino, Sáez, 1973). Solution A contained 3 g of $(NH_4)_2SO_4$, 0.1 g of KCl, 0.5 g of K_2HPO_4, 0.5 g of $MgSO_4H_2O$ and 0.01 g of $Ca(NO_3)_2$, this mixture of basic salts was diluted in 700 ml of distilled water and autoclaved at 120 °C for 15 minutes. Solution B was prepared with 44.8 g of $FeSO_47H_2O$ in 300 ml of distilled water. The ferrous sulfate heptahydrate ($FeSO_47H_2O$) was the energy source chosen for the adaptation process, but being susceptible to oxidation at high temperatures, it was sterilized by amphiphilic filtration in a Millipore membrane of 0.22 µm opening diameter. Then, both solutions were mixed in a microbiological safety cabinet to complete the preparation of the liquid 9K medium. 9K culture medium was poured in Erlenmeyer flasks and pH adjusted to either 2 or 1.5 with 10N H_2SO_4; then cultures were inoculated with 1 ml of water from the mine and others with 8 grams of pulverized rock. They were finally incubated in the dark at 35 °C, performing successive plantings in fresh media every 96 hours for 16 days.

In phase 2, the final objective was to get all the consortia to release heavy metals mainly present in the WPCBs, so it was necessary to gradually reduce the iron sulfate content in the medium and increase the content of WPCB (Chen, 2015). Finally, all cultures were terminated in Erlenmeyer flasks containing 90 ml of 9k medium without solution B, but with 20 g of WPCB. The process of this second adaptation was performed once all the cultures complied with the first phase. Bioassays were reproduced in triplicate for each temperature, pH and stirring conditions, in order to establish the incidence of these variables on the results.

Experimental design included four experimental manipulated variables: origin of bacteria consortia (mine water, mine rock, and WPCBs); pH (1.5 and 2.0); temperature (28 °C and 32 °C); and agitation speed (0 and 150 rpm). Response variables included concentration of metals in culture liquors.

2.4. Microbial growth kinetics

After completion of the second adaptation phase, all cultures were inoculated to fresh 9K (without ferrous sulfate) media and 20 g of WPCBs. From that moment, every 24 hours during the 4 days following the inoculation, aliquots of 100 µl of each bioassay were taken and were deep - seeded in petri dishes with 9k medium gelled with agar - agar (Merck). Finally, counting of colony forming units (CFU) was performed to establish the growth kinetics of the cultivable chemiolitotrophic microorganisms in the different bioassays studied. Likewise, the Gram staining technique was used from cultures broths, facilitating their observation and morphological identification (Arias et al., 2013). pH readings were performed on all assays for 24 hours until the end of the assay.

The determination of the number of viable colonies in each of the bioassays cultured under the different conditions was performed by the formula:

$$CFU/_{ml} = \frac{No.of\ colonies\ per\ plate\ \ x\ dilution\ factor}{ml\ of\ sample\ sown} \quad (1)$$

2.5. Kinetics of metal leaching

In the same cultures where the growth kinetics was determined, the leaching kinetics of the metals that could have passed into a soluble form from the WPCBs immersed in the cultures was evaluated. At 24, 48, 72 and 96 hours, 100 µl aliquots were taken from each culture, prepared with 100 µl of Ga 1000 mg/l standard in Eppendorf microtubes, centrifuged at 3000 rpm for 30 seconds and then tested for quantify the metals present in these liqueurs by X-Ray fluorescence (S2 Picofox, Bruker) (Marguí, E. et al., 2010).

3. Results and discussion

Multiple investigations have shown that several metals can be efficiently leached using *A. ferrooxidans* in shake flasks (Mejía, E., 2011; Gómez, J., 2005; Menadier, 2009). However, there is insufficient data on the use of microbial consortia and even leaching of heavy metals from WPCBs and their potential for commercial exploitation. In order to find the behavior of new consortia in the bioleaching of WPCBs, the growth of the isolated consortia in the presence of these under different cultivation conditions was analyzed for comparative purposes. In addition, the leached concentrations of the metals present in the WPCBs immersed in the different cultures were measured.

3.1. Kinetics of growth

Fig. 1 shows the growth of bacterial colonies in the course of time. Similar behavior was showed for all cultivated consortia. However, it was also clear the influence of the studies variables: microorganisms from acid mine waters were those that had the highest growth, followed by cultures inoculated with mine rocks and finally cultures isolated from WPCBs; likewise, the cultures that reported the highest growth were those that had agitation; cultures grew more at 32 °C than at 28 °C, as well as cultures at pH 2, compared to those at pH 1.5. This information is crucial for optimizing parameters in future research.

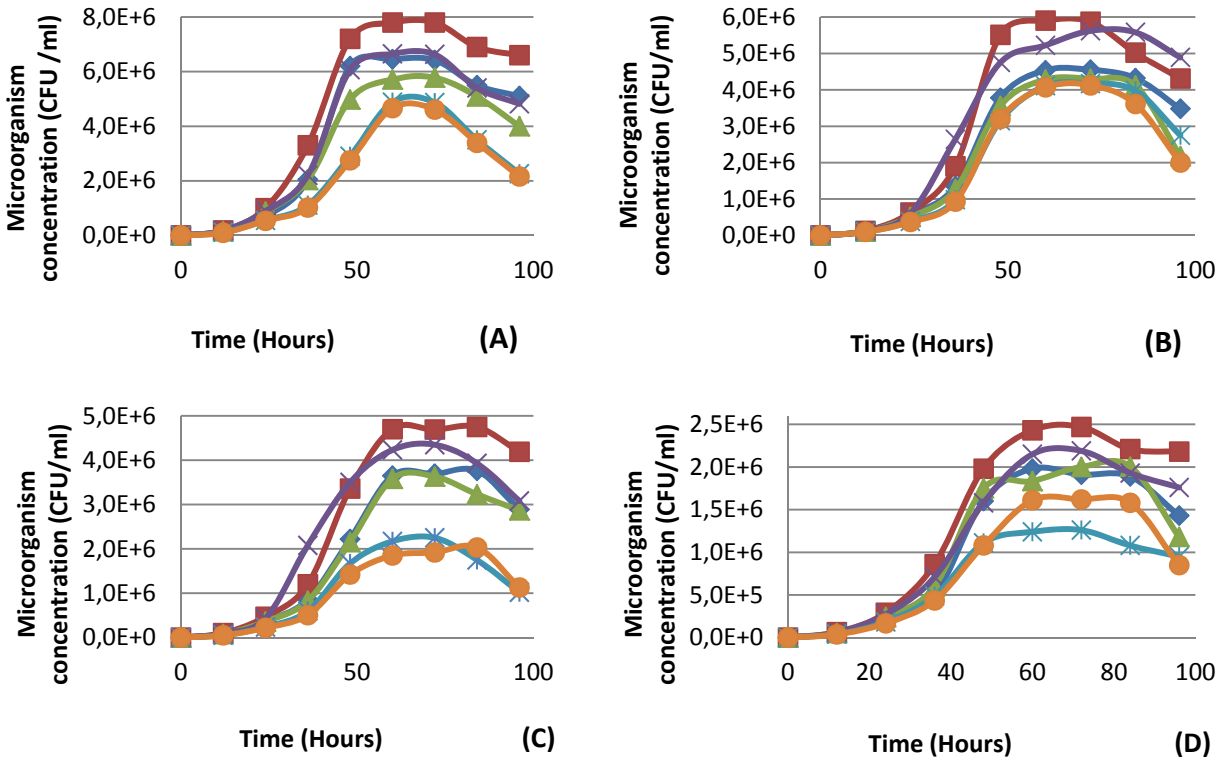

Fig. 1. Microorganism concentration (CFU/ml) vs time for the different strains and conditions of study. (A) 32 °C, 150 rpm; (B) 28 °C, 150 rpm; (C) 32 °C, 0 rpm; (D) 28 °C, 0 rpm. **Conventions:** Consortia isolated from mine water, initial pH 1.5 ( ); Consortia isolated from mine water, initial pH 2.0 ( ); Consortia isolated from mine rocks, initial pH 1.5 ( ); Consortia isolated from mine rocks, initial pH 2.0 ( ); Consortia isolated from WPCBs, initial pH 1.5 ( ); Consortia isolated from WPCBs, initial pH 2.0 ( ).

3.2. Morphological identification by Gram staining.

After the adaptation stage, the presence of colonies with Gram negative bacillary morphology, as well as the presence of Gram positive cocci was observed. Also, observation was made on fresh cultures and filamentous hyphae and spores were frequently observed. In addition to microorganisms mentioned above, some filamentous structures were observed which grew in the presence of ferrous sulfate (Fig. 2). According to the aforementioned morphology, it is assumed that in the several cultures

the presence of *Acidithiobacillus s.p*, *Thiobacillus s.p*, *Leptospirillum s.p* and *pseudomonas s.p* genera is observed.

3.3. Turbidity and pH

In general, cultures containing acidic mine-water inoculants showed a more pronounced trend in color reversal, which gives evidence of iron-to-ferric ion oxidation and leaching of other metals, as well as the precipitation of some salts (Results not shown). Meanwhile, Computer cultures were the clearest bioassays, with a yellow ocher hue that allows inferring that these had the lowest concentration values for leaching. Likewise, a change in color was observed in the control culture due to the oxidation of some salts and sulfurous metals that precipitated over time.

For all the bioassays, a decrease in the acidity with time was observed. The initial pHs of the samples, either 2 or 1.5, increased to pH values slightly lower than 2.5 during the first 24 to 48 hours. Subsequently, the pH stabilizes in some bioassays for almost 50 hours (Fig. 3). After 50 hours, the pH again varies drastically, exceeding values of 3 in Fig. 3A, 3C, and 3D. The other bioassays, on the other hand, show a tendency to be constant from the 48 hours and are maintained during almost 30 hours followed before returning to suffer a much more pronounced decrease in the degree of acidity is referred, reaching pH above 3 also. An example of this behavior is shown in Fig. 3B. This final increase of the pH can favor the inhibition in the growth of the studied consortia.

In addition, in all the bioassays initially adjusted to pH 2 there was a marked tendency to maintain a new pH value almost constant once values of 2.5 were reached. It was also detected that the cultures with more sediments decreased and in turn suffered a reduction of acidity faster. However, in the cultures with pH 1.5 it was not possible to reach pH values of 2.5 in the middle of the process, but, very close values; on average the cultures were able to stabilize at a pH of 2.27, although later all crops would lose acidity.

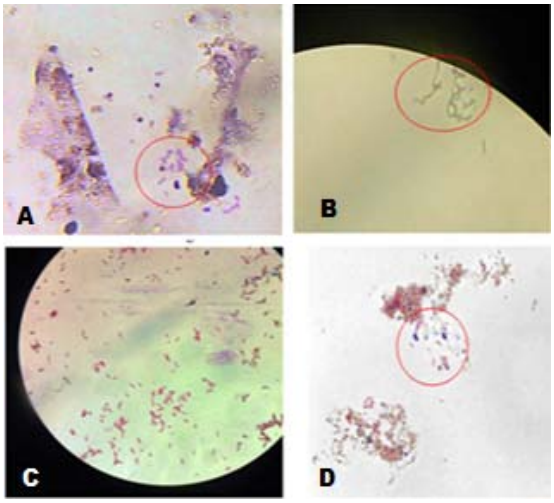

Fig. 2. Microscopic view of microorganisms present in bioassays in 9k cultures. A: Gram negative bacilli and Gram positive bacilli (isolated WPCBs inoculum). B: Hyphae and spores (isolated WPCBs inoculum). C: Gram negative bacilli (inoculum from water of mining action). D: Gram negative bacilli and Gram positive bacilli (inoculum of rock of mining action).

The initial increase in the pH of the bioassays could be due to factors such as: (i) The presence of carbonates and silicate phases in contact with the leaching solution generates acid consumption (Mejía et al., 2011); (ii) The consumption of protons generated mainly by the monosulfide solution (Ballester, 2005). (iii) The oxidation of Fe^{2+} present in the solution and (iv) the microbial consortia used (Özkaya et al., 2007). In our case, the initial pH increase in all bioassays is due to the presence of carbonate compounds, silicates, some metal oxides and alkaline substances belonging to the integrated circuits of discarded computers (Chen et al., 2015) immersed in each culture (about 29% silica), generating a consumption of hydrogen protons, thus contributing to the reduction of acidity during the first hours. This generalized increase in the pH of all cultures was able to inhibit the adequate growth of the

microorganisms and thus delay their adaptation, which is in agreement with the work of Meruane and Vargas (2003), and Mejía *et al* (2011). However, all the bioassays reached a stability in pH towards the middle of the process (Fig. 3), due to the production of H_2SO_4 by the microbial action result of the oxidation of ion Fe^{2+}, which suggests a neutralization of carbonates, since these minerals, in the presence of sulfuric acid, are neutralized to form gypsum and other types of sulfates depending on the chemical composition of the carbonates (Márquez, 1999) and proton release related to the hydrolysis and precipitation of the generated ferric iron (Ye, et al., 2017). In this sense, the ideal condition for the exponential growth of the microorganisms of each bioassay was generated from the regulation and stabilization of pH (Rodríguez, 1997).

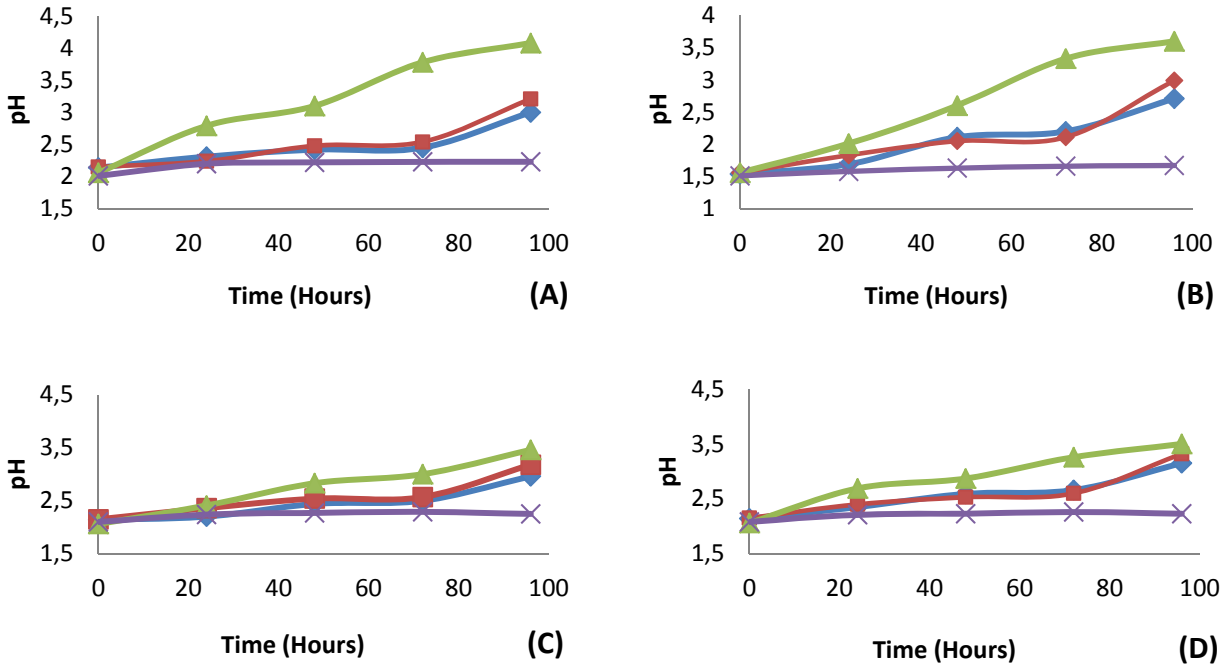

Fig. 3. pH vs time for cultures at: (A) 32 °C, pH 2.0, 150 rpm; (B) 32 °C, pH 1.5, 150 rpm; (C) 28 °C, pH 2.0, 150 rpm; (D) 32 °C, pH 2.0, 0 rpm. **Conventions**: Consortia isolated from mine water ( ); Consortia isolated from mine rocks ( ); Consortia isolated from WPCBs ( ); Control culture ( ).

Tolerance levels of toxic ions could have been the main cause for which the microbial population was decreasing after 96 hours (Bryan, CG et al., 2015), and as a consequence a new increase in pH. It has been suggested that the metals contained in the e-waste play the largest role in microbial inhibition (Ilyas et al. 2010). The activity of bacteria could be inhibited immediately by addition of elemental metal ions how Cu^{2+}, Cr^{3+}, Ni^{+}, and Zn^{2+} (Bryan et al., 2015) and all these ions were reported in our experiments. Additionally, PCBs were alkaline in nature. As time passed, more acid was needed to neutralize the alkaline substance in the PCBs and maintain an optimum pH for the microorganisms (Wu, W et al. 2018). This adjustment was not made in the present study, due to the interest to evaluate to what extent the microorganisms were able to stabilize their environment, because in nature there are no such adjustments. Somewhere else, precipitates cover the surface of the substrate preventing bacterial action. Consequence of this, after 96 hours, the pH continued to rise, also contributing to the inhibition of microbial growth. As a future perspective, precipitation reactions in leaching processes could be restricted under a constant pH, maintained by adding dilute sulfuric acid. Therefore, the cycle process (Fe^{2+} - Fe^{3+}) can be maintained and promote the recovery of various metals (Chen et al., 2015).

3.4. Kinetic of leaching of heavy metals

It is important to note that the assumption that aqua regia's extraction conditions will allow more metal dissolution rather than microorganism oxidation is made and, thus, results obtained by chemical

lixiviation were taken as the maximum lixiviation limit. This aqua regia procedure is not expected to lixiviate all metals and whether the microorganisms manage to lixiviate the metals in a higher proportion than the chemical treatment this will be reflected in an yields higher than 100%. One of the objectives of the present investigation was to characterize mainly the heavy metals leached as a product of the processes that allow the growth of the consortia studied. Results of the initial analysis of metals present in the WPCBs of desktop computers are presented in table 1 after the identification of each of them through the chemical analysis of TXRF.

These values were used as reference for the determination of the leaching yield of each of the metals. Thus, to perform the calculations of yield of all the leached metals the following equation was used:

$$\% Yield = \frac{S}{E} x100 \quad (2)$$

Where, *S* is the concentration of the leached metal at the end of the process and *E* the initial concentration of the metal.

Table 1. Metal distribution of metals within WPCBs Intel DDR3, as obtained with extraction with aqua regia.

Element	Concentration (mg/l)	Relative amount (%)
Ca	289.5	2.036
Cr	17.8	0.189
Mn	31.86	0.336
Fe	5830	61.517
Ni	192.96	2.036
Cu	717	7.566
Zn	667	7.038
Br	2.93	0.031
Sr	0.766	0.008
Sn	1756.33	18.532
Ta	4.056	0.043
Au	0.903	0.010
Pb	55.23	0.583
Os	6.03	0.064
Pt	2.63	0.028

Among the metals that were leached with the help of microbial action, the toxic heavy metals, lead, nickel, chromium, and osmium were extracted. In Fig. 4, the leaching kinetics of nickel, lead, and chromium metals, respectively, are shown as examples. Table 2 shows the final percentage of leaching of these metals in all the tests as well as other metals extracted during the process. Nickel was the only heavy metal that leached in all the bioassays studied. Unlike the lead that was favored more at 32 °C, the higher values of this metal were favored to the conditions of temperature of 28 °C and agitation of 150 rpm. The leaching of nickel under these conditions was much higher, compared to the rest of the bioassays, so much that it exceeded values of 50 mg /l (Fig. 4A).

Lead concentration had values above 10 mg/l in all cultures with agitation, obtaining the maximum value of 16.9 mg/l corresponding to the inoculated bioassay of microorganisms from mine water, adjusted to initial pH of 2 and under temperature conditions of 32 ° C and 150 rpm of agitation; on the other hand, in the cultures without agitation there was no significant extraction, having values even lower than 5 mg/l, which allows to infer that lead leaching improves under agitation conditions (Fig. 4B). For chromium, the best results were obtained for agitated cultures. The highest values for chromium leaching were found in cultures at 32 °C, however, there were more cultures that presented chromium leaching at 28 °C than at 32 °C (Fig. 4C). Besides, during the 4 days of monitoring the leaching other metals, including Fe, Ca, Zn, Mn, Cu, Ta, Pt, and Au was detected. Also presented in the readings were elements such as Si which is part of the structure of integrated circuits and Br which is used in brominated parts (Table 2). The presence of Os occurred only in the liquors of the bioassays inoculated with strains of water and rock from mines. The importance of the recovery of this metal is that it is one of the scarcer elements found in the earth's crust at a concentration of 0.0015 mg/kg.

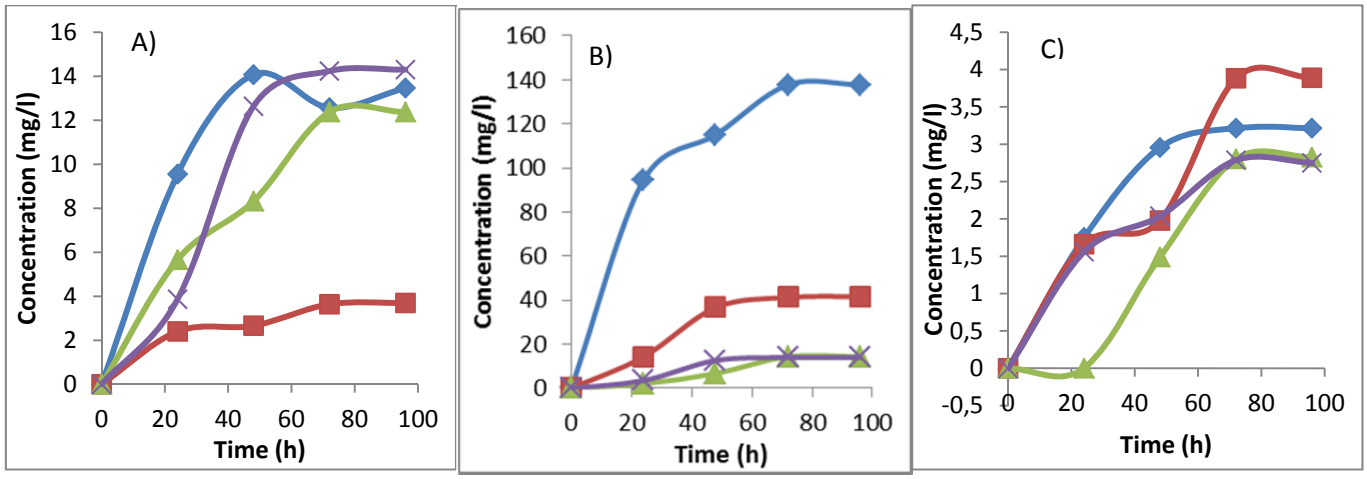

Fig. 4. Metal concentration in leached liquors of WPCBs by studied consortia: A) Pb; B) Ni; C) Cr. **Conventions**: Consortia isolated from mine rocks, pH 2.1, 28 °C, 150 rpm ( ); Consortia isolated from mine water, pH 1.5, 32 °C, 150 rpm ( ); Consortia isolated from mine water, pH 2.1, 28 °C, 150 rpm ( );Consortia isolated from mine rocks, pH 1.5, 32 °C, 150 rpm ( ).

Although not shown in Table 2, gold was only leached by the inoculated culture with strains from mine water, initially adjusted to pH 1.54, 32 °C and 150 rpm. Under these conditions the gold recovery was 91.144%. For the case of platinum, this was only solubilized in the liquor for the bioassays inoculated with strains from mine water at pH 2.15 and strains of mine rocks at pH 1.58; Both bioassays at 28 °C and without agitation, achieving a recovery of this metal of 8.677% and 21.835%, respectively.

Control cultures were used as reference for all bioassays. In the case of lead, the isolated bioassays in water and rock mining products reported higher leaching data for lead than the controls, which, although they showed a positive response to lead, in general, the behavior of leaching in These were very similar to those reported by the cultures isolated from WPCBs, which were much lower than the cultures mentioned above. Although control cultures that showed a positive response to lead showed in general a leaching-like behavior presented by the asylum cultures from WPCBs. As for chromium, it is important to note that neither control cultures nor cultures with isolated inoculants from WPCB reported chromium leaching under any condition.

The similar behavior in the control cultures and cultures inoculated from WPCBs suggests that the results for the leaching of the metals in the latter were more product of reactions, due to the consumption of Fe^{2+} ion than of the same presence of the microorganisms, although it is necessary to fathom in this aspect because there was evidently microbial growth, prevailing in these the presence of heterotrophic fungi and microorganisms of which already has been reported in the literature are able to grow under high concentrations of metals (Morales, 2008), although the results for the present study were not the most encouraging. Theoretically, bacterial bioleaching processes can continue until all the substrate is converted into product, but in closed systems as happened with the bioassays of the present study, the accumulation of products at high levels can cause toxicity to microbial cultures (Ahoranta et al., 2016). Thus, inhibition of microbial activity by some component of the WPCBs would compromise bioleaching process (lixiviate production) and reduce the rate of metals dissolution (Bryan, C, et al. 2015) and has been thought to be the major reason of low-efficient leaching (Yang et al., 2014). Another factor that could influence bacterial activity, although not from a biological point of view, but which determines the effectiveness of the bacterial attack, is the size of the mineral particles, since it determines the surface area exposed to the action of microorganisms. Usually, the bleaching performance increase with lower particle size (Muñoz, 1992). However, the grinding of these materials is a very costly process that further hinders the reduction of particles and their large scale realization at the present time.

Table 2. Extraction yields of bioleaching at the studied conditions.

Temperature and agitation	Initial pH	Inoculum	% Ni	%Pb	%Cr	% Fe	% Ca	%Mn	%Zn	%Cu	%Os	%Ta
32°C, 150 rpm	1.54	Acid waters	21.40	6.57	0	68.67	0	39.38	20.84	0.51	27.01	59.40
	1.56	Mine rock	7.250	25.56	15.56	55.03	0	42.67	4.583	0.11	12.37	57.27
	2.14	Acid waters	1.94	30.61	21.69	60.49	66.51	0	11.489	0.36	0	28.76
	2.15	Mine rock	7.46	28.55	0	71.19	80.51	49.37	4.953	0.88	8.23	53.16
	1.56	PC parts	2.46	0	0	34.30	64.94	0	0.825	0	0	0
	2.06	PC parts	2.06	11.75	0	40.41	30.65	0	0.925	0	0	0
	1.55	Control	2.01	0	0	39.91	32.11	0	1.092	0	0	0
	2.03	Control	1.85	12.36	0	36.99	62.74	0	0.726	0	0	0
28°C, 150 rpm	1.55	Acid waters	43.61	19.49	0	69.48	49.18	48.53	8.881	0.59	0	89.56
	1.54	Mine rock	42.85	5.78	0	68.21	43.00	45.92	13.658	6.31	14.53	73,04
	2.14	Acid waters	7.46	22.39	15.17	61.41	0	0	12.324	0.45	0	0
	2.1	Mine rock	71.41	22.75	17.96	66.54	50.25	43.72	7.096	0.21	10.16	88.74
	1.99	PC parts	1.83	10.11	0	19.63	36.47	43.09	0.618	0.17	0	0
	1.54	PC parts	1.70	11.46	0	31.41	53.52	0	0.775	0	0	0
	1.54	Control	1.59	10.85	0	33.49	40.04	0	1.11	0	0	0
	2.13	Control	1.68	9.96	0	19.06	35.11	39.68	0.53	0.18	0	0

4. Conclusions

Bioleaching of heavy metals such as iron, calcium, zinc, manganese, copper, osmium, tantalum, platinum and gold from WPCBs using different microorganisms Sources was demonstrated. Experimental results show that the rate of dissolution of the metals can be regulated by controlling parameters such as temperature, pH, stirring speed and density of extract, which facilitate bacterial growth. Likewise, the adaptations for successive reseeding showed good results on the different consortia.

It is noteworthy that the bioleaching kinetics of the different metals was affected by the interruption in bacterial growth, as well as by the formation of precipitates, the toxicity resulting from the increasing level of leached metals and the increase in the pH of the solutions. Therefore, as a future perspective, precipitation reactions in the leaching processes could be restricted under a constant pH, maintained by adding dilute sulfuric acid. Thus, the cycle process (Fe^{2+} - Fe^{3+}) can be maintained and promote various metals recovery.

Also, fully understanding the optimum solids concentration, particle size and pH values may influence how the bioleaching is carried out and how leaching of one metal selected over another would be favored. It is suggested the use of mixtures of microorganisms, which by symbiotic phenomena can accelerate reactions and achieve richer solutions.

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