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Bioleaching of electronic waste

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ABSTRACT: Increase in advanced electronic technology leads to environmental issues related with its disposal. Electronic waste i.e., video card and random access memory were used for studying extraction of precious metals using *Paenibacillus* sp. Metal contaminated soil was used for the isolation of exopolysaccharide producing strains. The isolate was identified as *Paenibacillus* sp. based on morphological, biochemical tests and 16S rRNA sequencing. Metal content analysis of soil and e-waste was carried out using X-ray Fluorescence spectroscopy. The vanadium element was more in the soil sample which was 0.487 mg/g and in electronic waste sample copper content was more which was 250 mg/g. *Paenibacillus* sp. produced capsule which was observed under bright, dark field and phase contrast microscope. Scanning electron microscopy was done for the study of morphological changes of exopolysaccharide producing *Paenibacillus* sp. in chitin broth and on chitin agar medium with and without e-waste. The Fourier Transform Infrared Spectroscopy analysis of exopolysaccharide produced by *Paenibacillus* sp. grown on chitin agar and chitin agar with e-waste showed presence of different functional groups. The one step and two step bioleaching experiments were carried out for testing efficacy of biomass on metal leaching. *Paenibacillus* sp. showed its potential for the extraction of precious metals viz., gold, silver and copper from electronic waste. *Paenibacillus* sp. recovered gold (0.001%), cadmium (45%), copper (50%), iron (46%), manganese (88%), palladium (56.9%) and zinc (87.12%) by two step fermentation. The study is useful for the bioleaching of precious metals from electronic waste.

Keywords: Exopolysaccharides, Microbial extraction, *Paenibacillus*, Bioflocculation, Eco-friendly.

INTRODUCTION

Advancement in technology leads to progressive use of electrical and electronic equipment which leads to generation of electronic waste (e-waste). Around 50 million tons of e-waste generates worldwide. The disposal of e-waste is a major challenge as it contains many toxic elements viz., lead, mercury, arsenic, cadmium, selenium, chromium, etc. Management of e-waste has now become a serious concern in developed as well as developing countries. Precious metal content, energy requirement and pollution control measures enable to recycle the waste rather than using it for landfilling purposes. For the recovery of metals there are number of processes viz., mechanical separation, pyrometallurgical, hydrometallurgical and bio-hydrometallurgical. These processes have

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some disadvantages viz., high energy requirement, insufficient recovery of precious metals, formation of toxins i.e., dioxins and furans (Li et al., 2007), cyanide causing environmental pollution. Recent interest in the biotechnology is to treat the waste and low grade ores for recovery of precious metals. Bacterial activities like bioleaching (Pradhan & Kumar, 2012), bioprocessing, biobeneficiation, biosurface modification, bioflocculation and bioremediation are applicable for extraction of precious metals and scarce elements. Bioleaching is now the most important emerging technology for various metal extractions from e-waste. The bioleaching process has advantage as it is eco-friendly process. The recovery of metals can be enhanced by bioleaching process. This is seen in case of copper and gold mining where low grade ores are biologically treated to extract various valuable metals (Sajjad et al., 2019). Bioleaching can be done by one and two step process. In biological leaching, natural agents transform metals and it is one of the efficient biohydrometallurgical processes. *Chromobacterium violaceum* has been reported in the recovery of gold (Pham & Ting, 2009). There is study where the bacteria isolated from the marine sponge of *Hymeniacidon heliophila* found to be used for copper recovery from e-waste (Rozas et al., 2017). The bioleaching of gold, copper and nickel from waste cellular phone PCBs and computer goldfinger motherboards by two *Aspergillus niger* strains has been reported (Madrigal-Arias et al., 2015).

There is a report on bioleaching of copper from waste PCBs by bacteria-free cultural supernatant of *Leptospirillum ferriphilum* and *Sulfobacillus thermosulfidooxidans* (Wu et al., 2018). Also, study is there on bioleaching of gold from PCBs of mobile phones by *A. niger* in a culture without agitation and glucose as carbon source (Argumedo-Delira et al., 2019). Other than the toxic elements; the e-waste contains various significant amounts of precious metals also like gold. The precious metals are present in dozens of times the metals present in high-grade ore. In case of gold, the natural gold ores contain 0.5-13.5 g/ton, and e-waste contains 10-10,000 g gold/ ton (Cui & Zhang, 2008; Pham & Ting, 2009). Thus, e-waste is important economic source for the recovery of precious metals.

The work here describes the study of *Paenibacillus* sp. for the recovery of metals using the one and two step bioleaching process. This bioleaching process will be economical and very easy and help to recover many precious metals.

MATERIALS AND METHODS

Sampling and analysis of the soil and e-waste for heavy metal content

Soil was collected from Tukai hill located in Hadapsar, Pune, Maharashtra, India [18.482165 latitude and 73.944938 longitudes]. Soil was found to contain heavy metals viz., chromium, vanadium, copper, zinc and barium in high concentration. Many bacterial species produce exopolysaccharides (EPS) in response to these heavy metals (Nocelli et al., 2016). Hence, the heavy metal soil was selected, packed in polycarbonate bag, labeled and used for the isolation of EPS producing bacteria. The pH of the soil sample was measured using the pH meter (Karastogianni et al., 2016) and humidity by hygrometer (Kronenberg et al., 2000).

Different kinds of e-waste like RAM and mother board were collected from area called Lonikalbhor, Pune, Maharashtra, India. Pulverization or grinding of e-waste was done to particle sizes ≤150 μm [i.e., standard test sieves as per IS 460: 1962] using the motorized grinding machine. The soil and e-waste samples (1 g each) were analyzed for the metal content using X-Ray Fluorescence [XRF] Spectrometry (Spectroxepos Ametek, Germany).

Isolation and identification of EPS producing isolate

The enrichment and isolation of EPS producing organism was done on collidal chitin agar medium [colloidal chitin: colloidal chitin: 0.4%, magnesium sulphate: 0.01%, dipotassium phosphate: 0.01%, potassium dihydrogen phosphate: 0.01%, distilled water: 100 mL] by serial dilution of the soil sample. EPS producing colonies were selected. The isolates were grown on nutrient agar [NA] and chitin agar plate containing 10 to 100 ppm concentration of copper sulphate, silver nitrate, mercuric chloride and nickel chloride. The isolate which was found to produce EPS and tolerance to all metals at 100 ppm was selected and used for the bioleaching studies. Morphological characteristics, biochemical tests viz., nitrate reduction (Buxton, 2011), indole (Darkoh et al., 2015), methyl red [MR] (Haque & Sao, 2015), citrate utilization (Haque & Sao, 2015) and hydrogen sulphide [H_2S] production (Tambekar et al., 2007) were studied for the identification of EPS producing bacteria. The enzymes production viz., catalase, oxidase, urease, phenylalanine deaminase, gelatinase, chitinase, phosphatase, caseinase, amylase, pectinase and lecithinase were also performed for the identification of the isolate (Aneja, 2001). The isolate was confirmed by doing 16S rRNA sequencing.

Microscopy study of capsule production and growth of *Paenibacillus* sp. in chitin broth and on chitin agar medium with and without e-waste

Capsular polysaccharide was assessed by Maneval's staining method (Cornelissen et al., 2011). For capsule staining, a loopful of culture was taken on a clean glass slide. A drop of 1% congo red was added and spread gently to make a smear and air dried. The slide was flood with Maneval's stain and kept for 2 min. After 2 min, the excess stain was removed and the slide was air dried. The capsule staining was observed using bright and dark field [Olympus Inverted Compound Microscope] and phase contrast microscopy [Nikon Ti-S Inverted Epifluorescence Microscope with Phase Contrast]. *Paenibacillus* sp. was grown on chitin medium containing 1g% crushed e-waste. Inoculated plate and broth were incubated at room temperature for 72 hr. Microbial growth was scrapped from agar surface and kept in oven at 40 ^{0}C for drying. Fermented chitin broth with 1 g% e-waste of *Paenibacillus* sp. was centrifuged for recovery of pellet containing microbial biomass and e-waste. Pellet was subjected for drying in oven at 40 ^{0}C. The dried samples viz., e-waste sample; *Paenibacillus* sp. grown in chitin broth; on chitin agar medium with and without e-waste were used for the scanning electron microscope (SEM) [SEM- Jeol, Tokyo, Japan] studies.

Fourier Transform Infrared Spectroscopy of EPS produced by *Paenibacillus* sp.

Characterization of EPS was done by Fourier Transform Infrared Spectroscopy [FTIR] [Perkin Elmer, USA] for which 48 h growth of *Paenibacillus* sp. was used. Functional groups in EPS were determined using ATR-FTIR [Perkin Elmer, USA].

Studies on bioleaching of e-waste using *Paenibacillus* sp.

The one step and two step bioleaching of metals by *Paenibacillus* sp. were formed as per the method of (Willner & Fornalczyk, 2013) and (Kumar, 2014) using colloidal chitin broth amended with 1 g% w/v powdered e-waste such as video cards and random access memory [AM]. Incubation was done for 10 days at room temperature for one step leaching process. In two step leaching process, 1 g% w/v powdered e-waste was added to 10 days fermented chitin broth of *Paenibacillus* sp. After incubation of 10 days, the media was centrifuged at 3000 rpm for 10 min and filtered through Whatman filter paper No.1. The amount of metals leached was analyzed by Atomic Absorption Spectrophotometer [AAS] [Varian

SpectraA, Germany]. The e-waste samples were also analyzed for metal analysis content by the chemical leaching method (Ilyas et al., 2007); AAS and Inductively Coupled Plasma Mass Spectrometer [ICP-MS] [AGILENT 7800 ICP-MS] for the one and two step bioleaching process.

Table 1. XRF spectrometric analysis of soil sample

Symbol	Elements	Concentration (mg/g)
Cd	Cadmium	< 0.002 ± 0.00
Co	Cobalt	0.050 ± 0.00
Cr	Chromium	0.270 ± 0.01
Cs	Cesium	< 0.004 ± 0.00
Cu	Copper	0.163 ± 0.00
Mo	Molybdenum	0.0093 ± 0.00
Ni	Nickel	0.113 ± 0.01
Sb	Antimony	0.0025 ± 0.00
Se	Selenium	< 0.005 ± 0.00
Sn	Tin	0.0085 ± 0.00
V	Vanadium	0.487 ± 0.01
Y	Yttrium	0.026 ± 0.00
Zn	Zinc	0.110 ± 0.00
Zr	Zirconium	0.151 ± 0.00

Data are represented as mean ± SD

Table 2. XRF analysis of the e-waste

Symbol	Elements	Element content (mg/g)
Ag	Silver	0.0203 ± 0.00
AlO	Aluminium	10.8 ± 0.01
Ba	Barium	0.02913 ± 0.00
Bi	Bismuth	0.0052 ± 0.00
CaO	Calcium	10 ± 0.02
Ce	Cerium	0.0606 ± 0.00
Cl	Chlorine	0.245 ± 0.00
Cr	Chromium	0.01723 ± 0.00
Cs	Caesium	0.0245 ± 0.00
Cu	Copper	250 ± 0.04
FeO	Iron	65.2 ± 0.02
I	Iodine	0.0790 ± 0.00
MgO	Magnesium	12.1 ± 0.02
MnO	Manganese	0.9 ± 0.01
Mo	Molybdenum	0.0373 ± 0.00
NaO	Sodium	0.1 ± 0.00
Rb	Rubidium	0.0806 ± 0.01
S	Sulphur	1.025 ± 0.00
TiO	Tin	4.8 ± 0.00
V	Vanadium	0.054 ± 0.00
Y	Yttrium	0.0713 ± 0.01
Zr	Zircon	0.03712 ± 0.00
Cd	Cadmium	0.001 ± 0.00
Ni	Nickel	13.244 ± 0.02
Pb	Lead	12.129 ± 0.02
Zn	Zinc	12.271 ± 0.01

Data are average of mean ± SD

RESULTS AND DISCUSSION

Soil and e-waste analysis for the metal content

The pH of the soil sample was 6.9 and humidity was found to be 68%. Element present in soil enhance its aggregation which is due to EPS production from bacteria or plant. The soil sample analysis was done by XRF Spectrometry is shown in Table 1. The vanadium element was found to be more which was 0.487 mg/g, followed by chromium which was 0.270 mg/g (Table 1).

The XRF analysis of e-waste showed the presence of various elements viz., Ag, Al, Ba, Ca. Ce, Cl, Cr, Cs, Cu, Fe, I, Mg, Mn, Mo, Na, Rb, etc. [Table 2]. The Cu content was more [250 mg/g] followed by Fe [65.2 mg/g] [Table 2]. The elements Cd and Bi were present in very minor amount which was 0.001 and 0.0052 mg/g respectively. The XRF analysis proves that e-waste is a source of many elements.

Isolation and identification of the EPS producing isolate

The total isolates obtained were 8, out of which only isolate No. 3 produced EPS. The EPS producing colonies were mucoid, slimy and showed string-forming appearance. Based on the morphological characteristics, the isolate was found to be Gram positive motile rod with the presence of a capsule. The isolate was methyl red [MR] positive [Table 3] and showed production of enzymes viz., oxidase, catalase, urease, chitinase, phosphatase, caseinase, amylase, pectinase and lecithinase [Table 4]. Based on the morphological characteristics, biochemical tests and 16S rRNA sequencing, the isolate was identified as *Paenibacillus* sp. [Accession No. JX280499.1]. EPS of *Acidithiobacillus ferrooxidans* has been reported to increase metal leaching capacity (Bellenberg et al., 2012). Bioleaching of metals is reported to take place by contact and non-contact processes. In case of contact bioleaching, capsular polysaccharide binds to the metals which results in dissolution of metals at microbe-mineral interface (Mitsunobu et al., 2016). The exact mechanism of this is still unknown. The growth of *Paenibacillus* sp. on chitin agar with 100 ppm copper sulphate is represented in Fig. 1. *Paenibacillus* strain RM, endophyte of *Tridax procumbens* has been reported to have resistance to metals like copper [750 ppm], zinc [500 ppm], arsenic [400 ppm] and lead [500 ppm] (Govarthanan et al., 2016).

Table 3. Biochemical tests

Biochemical tests	Result
Nitrate reduction	-
Indole	-
MR	+
Citrate utilization	-
H_2S	-

MR: methyl red, H_2S: hydrogen sulphide
+: Positive, -: Negative

Table 4. Production of enzymes

Enzymes	Result
Catalase	+
Oxidase	+
Urease	+
Phenylalanine deaminase	-
Gelatinase	-
Chitinase	+
Phosphatase	+
Caseinase	+
Amylase	+
Pectinase	+
Lecithinase	+

+: Positive, -: Negative

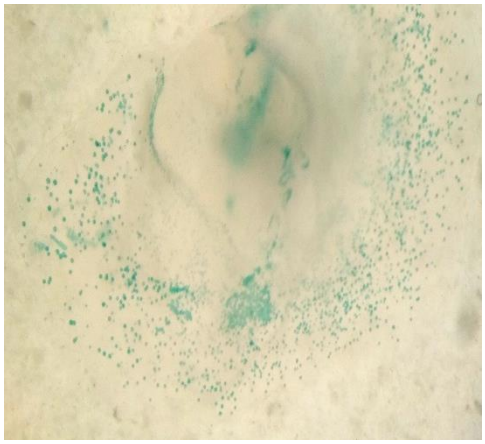
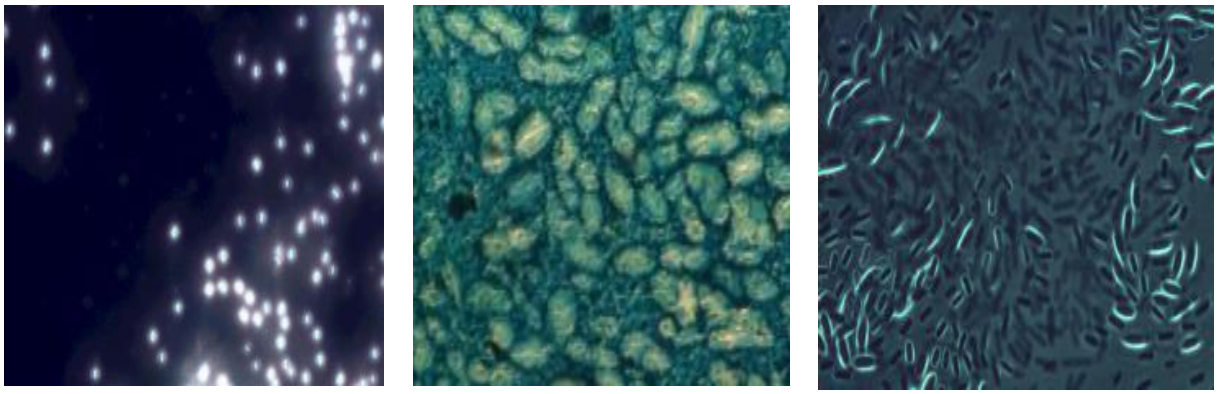


Fig. 1. Growth of *Paenibacillus* sp. on chitin agar with 100 ppm copper sulphate

Microscopy study of capsule and growth of *Paenibacillus* sp. in chitin broth and on chitin agar medium with and without e-waste

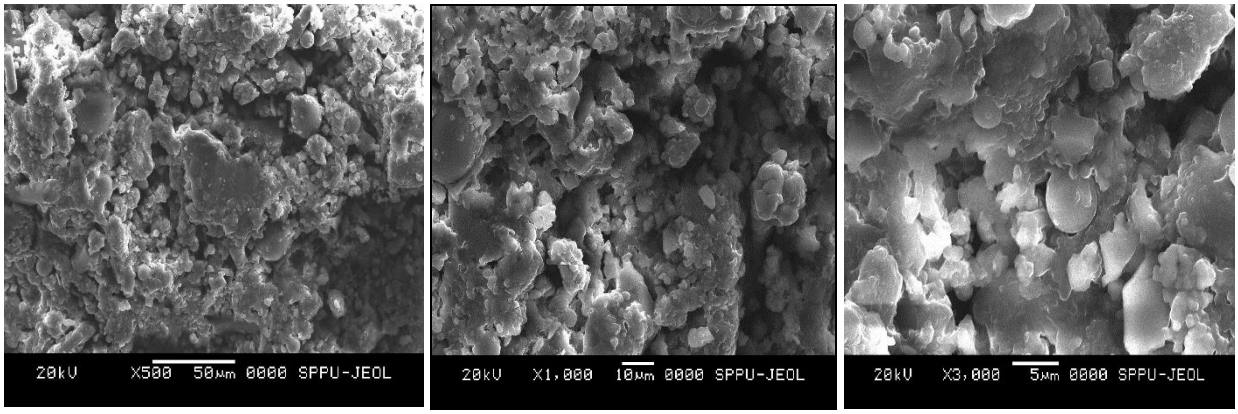
The presence of capsule of *Paenibacillus* sp. observed using dark, bright field and phase contrast microscope is shown in Fig. 2 a, b and c. The SEM images showed the polymorphic nature of e-waste observed under various magnification [Fig. 3 a, b and c]. EPS production was found to be reduced in the presence of e-waste. The

morphology of *Paenibacillus* sp. grown in chitin broth [Fig. 4 a, b and c], on chitin agar with e-waste [Fig. 5 a, b and c] and on chitin agar without e-waste [Fig. 6 a, b and c] showed varied growth of *Paenibacillus* sp.



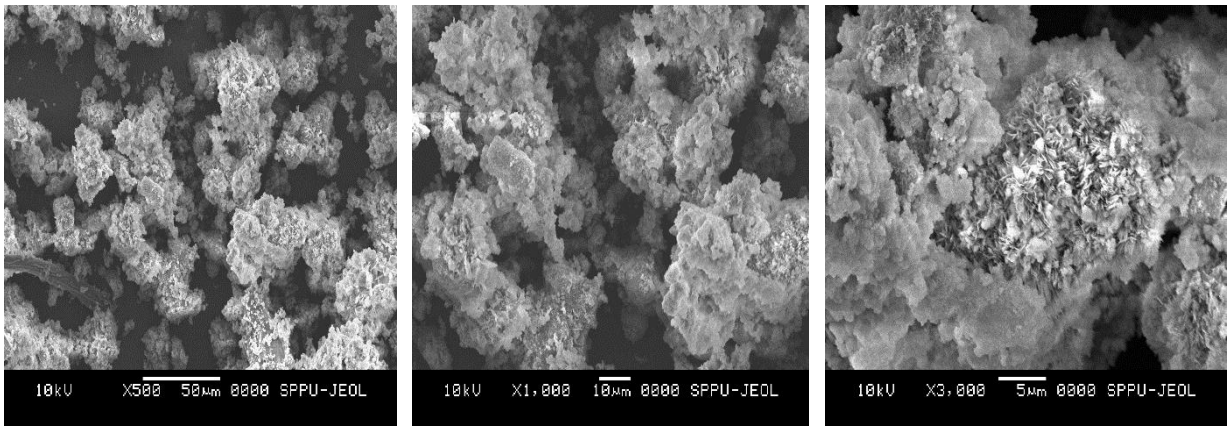
a b c

Fig. 2. Capsule of *Paenibacillus* sp. observed under a) dark field, b) bright field and c) phase contrast microscope respectively



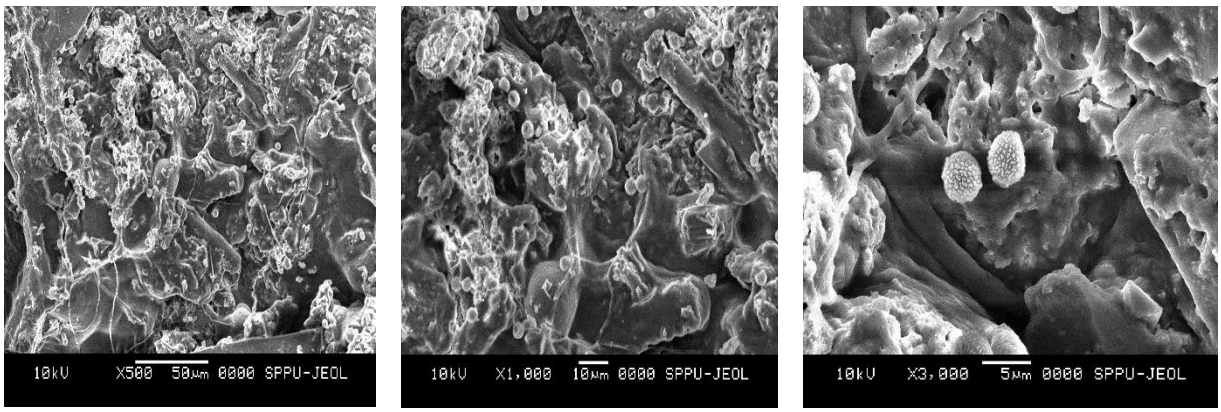
a b c

Fig. 3. SEM image of e-waste observed under a) X 500, b) X 1000 and c) X 3000 respectively



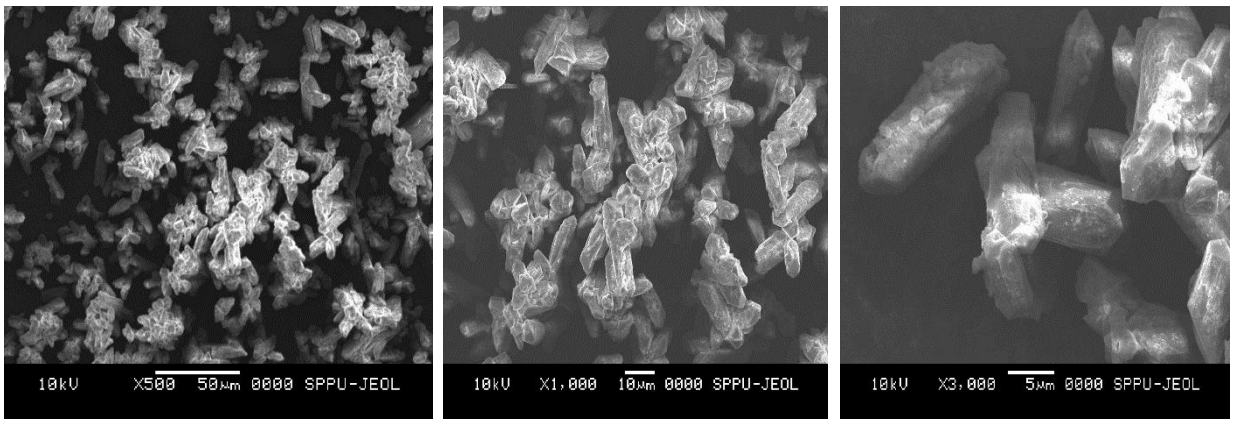
a b c

Fig. 4. SEM image of *Paenibacillus* sp. grown in chitin broth a) X 500, b) X 1000 and c) X 3000 respectively



a b c

Fig. 5. SEM image of *Paenibacillus* sp. grown on chitin agar with e-waste a) X 500, b) X 1000 and c) X 3000



a b c

Fig. 6. SEM image of *Paenibacillus* sp. grown on chitin agar plate a) X 500, b) X 1000 and c) X 3000

FTIR analysis of EPS produced by *Paenibacillus* sp.

The EPS produced by *Paenibacillus* sp. showed characteristic infrared absorbance patterns. More absorption bands were observed when *Paenibacillus* sp. was grown on chitin agar without e-waste [Fig. 7a]. *Paenibacillus* sp. grown on chitin agar in presence of e-waste also showed the absorption peaks [Fig. 7b] (Li et al., 2017; Naumann, 2000). The FTIR spectral analysis of EPS produced by *Paenibacillus* sp. grown on chitin agar and chitin agar with e-waste showed presence of different functional groups [Table 5]. The functional groups observed in case of *Paenibacillus* sp. grown on chitin agar were -OH group of H_2O, amide I and III group of proteins and -C-O-C group of polysaccharides. In case of *Paenibacillus* sp. grown on chitin agar containing e-waste, the functional groups observed were -OH group of H_2O, amide I group of proteins and -C-O-C group of polysaccharides [Table 5]. FTIR spectra showed that *Paenibacillus* sp. have adopted different biochemical features depending on the presence of nutrient material and heavy metals present in e-waste. Due to low penetration power spectral patterns generated using FTIR reflects the surface composition of the bacteria. There is a study where EPS is shown to be associated to soluble microbial by-product and aromatic compounds (Xia et al., 2019).

Table 5. FTIR spectral analysis of EPS produced by *Paenibacillus* sp. grown on chitin agar and chitin containing e-waste

FTIR spectral analysis of EPS produced by			
***Paenibacillus* sp. grown on chitin agar**		***Paenibacillus* sp. grown on grown on chitin agar containing e-waste**	
Peaks observed at wave number (cm^{-1})	Functional group	Peaks observed at wave number ($cm^{-1)}$	Functional group
3363.97	-OH group of H_2O	3340.30	-OH group of H_2O
2042.28	Nucleic acid weak band or amide I group	1534.45	amide I group
1621.58	Nucleic acid weak band or amide I group	1036.74	-C-O-C group of polysaccharides
1485.58	amide III group of proteins		
1292.76	-C-O-C group of polysaccharides		
1193.06	-OH group		

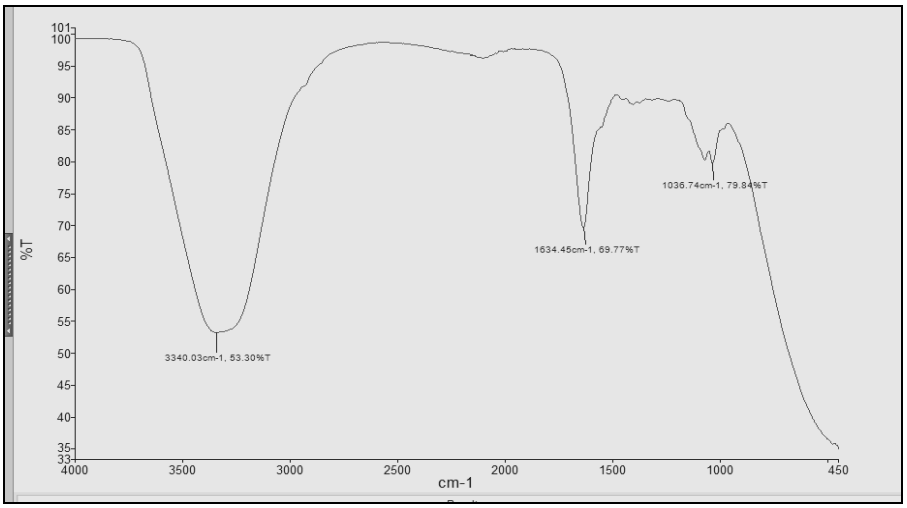


Fig. 7a. FTIR spectra of EPS produced by *Paenibacillus* sp. grown on chitin agar

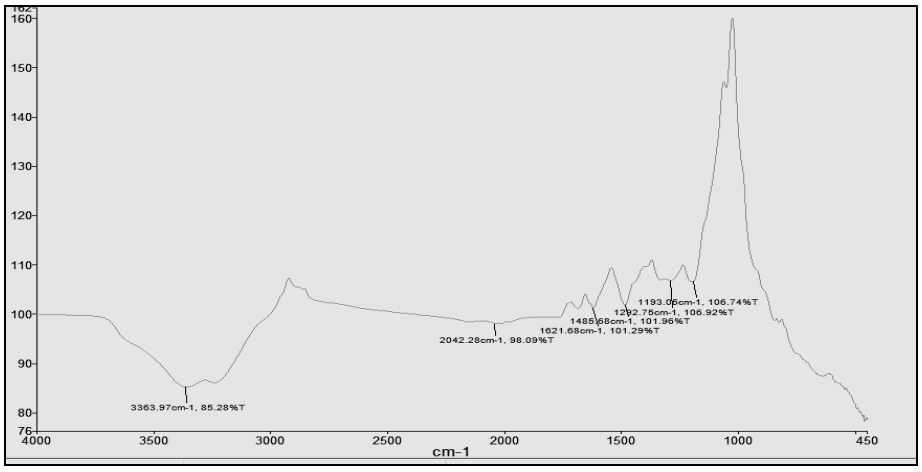


Fig. 7b. FTIR spectra of EPS produced by *Paenibacillus* sp. grown on grown on chitin agar containing e-waste

One and two step bioleaching process of e-waste by *Paenibacillus sp.*

The metals leached from e-waste [video card] using one and two step bioleaching processes by *Paenibacillus* sp. is shown in Table 6. Results obtained after one and two step bioleaching were compared with the metal concentration by chemical

leaching method [Table 6 and 7]. *Paenibacillus* sp. showed the potential to leach metals like cadmium, manganese, palladium and zinc from video card by both one step and two step leaching method. Recovery of cadmium and palladium was reported to be higher as compared to manganese and zinc. From RAM, *Paenibacillus* sp. recovered the metals like gold [0.001%], cadmium [45%], copper [50%], iron [46%], manganese [88%], palladium [56.9%] and zinc [87.12%] by two step fermentation. But for copper recovery, one step bioleaching was found to be effective as 70.45% copper was recovered. The metals leached from e-waste [RAM] using one and two step bioleaching process by *Paenibacillus* sp. is shown in Table 7.

The metals leached using one and two step bioleaching process by *Paenibacillus* sp. using AAS is represented in Table 8. This was compared with the chemical leaching method [Table 8]. Also, the metal analysis by ICP-MS of one and two step bioleaching process is represented in Table 9.

Similar results has been reported by *Streptomyces albidoflavus* TN10 which recovers Al [66%], Ca [74%], Cd [65%] , Fe [42%], Cu [68%), Ni [81%], Ag [56%] and Zn [82%] when grown at room temperature and pH 6.0 (Kaliyaraj et al., 2019).

Inductively coupled plasma mass spectrophotometer has been reported to be more sensitive than atomic absorption spectrophotometer as it could detect trace element also (Batsala et al., 2012). Hence, metal content were analyzed by both AAS and ICP-MS. Very poor leaching ability of strain was observed for precious metals in e-waste like gold and silver [10% in both one and two step bioleaching experiment].The metal analysis from e-waste using ICP-MS is shown in Table 9. According to the data obtained *Paenibacillus* sp. could leach metals like aluminum, copper, cadmium, nickel, zinc and lead more effectively than other metals in e-waste.

Work has been carried out on bioleaching from e-waste by *Chromobacterium violaceum* and *Pseudomonas* sp. (Pradhan & Kumar, 2012). Report is available on extraction of metals from e-waste by bacterial leaching where it describes the mechanisms in which the chemical reaction copper bioleaching from e-waste with the participation of *Acidithiobacillus ferrooxidans* bacteria (Willner & Fornalczyk, 2013). Study has been done on extraction of precious metals from e-waste using hydrometallurgical, pyrometallurgical and bioleaching process (Kavitha, 2014). Bio-oxidation mechanism has been reported in gold bioleaching of e-waste by cyanogenic bacteria (Pham & Ting, 2009). Bioleaching of copper from e-waste using *Acidithiobacillus* sp. has also been reported. *Acidithiobacillus ferrooxidans* and *Acidithiobacillus thiooxidans* have been studied in nickel bioleaching from PCBs (Mrazikova et al., 2014). There is also a report on extraction of metals from waste PCBs in solution containing copper sulphate and sulphuric acid (Yazici & Deveci, 2013).

Table 6. Leaching of metals from e-waste (video card) by *Paenibacillus* sp.

Symbol	Metals	Metals leached (mg/g of e-waste) in		
		One step bioleaching	Two step bioleaching	Chemical leaching
Cd	Cadmium	0.52 ± 0.01	0.345 ± 0.00	1.5 ± 0.00
Mn	Manganese	2.05 ± 0.00	2.400 ± 0.01	3.1 ± 0.00
Pd	Palladium	2.57 ± 0.00	4.2 ± 0.01	4.5 ± 0.01
Zn	Zinc	7.5 ± 0.01	12 ± 0.02	9.8 ± 0.02

Data are average of mean ± SD

Table 7. Leaching of metals from e-waste (RAM) by *Paenibacillus* sp.

Symbol	Metals	Metals leached (mg/g of e-waste) in		
		One step bioleaching	Two step bioleaching	Chemical leaching
Au	Gold	0.001 ± 0.00	0.001 ± 0.00	14 ± 0.03
Cd	Cadmium	2.14 ± 0.00	1.630 ± 0.00	2.5 ± 0.00
Cu	Copper	155 ± 0.03	110 ± 0.04	220 ± 0.03
Fe	Iron	3.48 ± 0.01	4.6 ± 0.01	10.0 ± 0.00
Mn	Manganese	1.87 ± 0.00	3.02 ± 0.00	3.6 ± 0.01
Pd	Palladium	1.275 ± 0.00	1.195 ± 0.00	2.1 ± 0.00
Zn	Zinc	6.7 ± 0.00	7.5 ± 0.02	8.6 ± 0.02

Data are average of mean ± SD

Table 8. Metal analysis from mixture of e-waste (video card and RAM) using atomic absorption spectrophotometer

Symbol	Metals	Metal content (mg/g)		
		One step bioleaching	Two step bioleaching	Chemical leaching
Al	Aluminium	135 ± 0.02	125 ± 0.01	141.50 ± 0.00
Ca	Calcium	51.4 0 ± 0.03	76.64 ± 0.01	588.50 ± 0.002
Cd	Cadmium	1.6 ± 0.00	2 ± 0.00	2.5 ± 0.00
Cr	Chromium	0.6 ± 0.00	0.04 ± 0.00	0.94 ± 0.00
Cu	Copper	575 ± 0.04	615 ± 0.04	1221.00 ± 0.02
Fe	Iron	140 ± 0.02	42 ± 0.00	363.90 ± 0.04
K	Potassium	296.8 ± 0.04	814.50 ± 0.03	884 ± 0.05
Mg	Magnesium	67.56 ± 0.00	39.64 ± 0.02	274.2 ± 0.01
Mn	Manganese	4 ± 0.00	3.8 ± 0.01	4.60 ± 0.00
Na	Sodium	84.51 ± 0.03	28.00 ± 0.00	102.30 ± 0.00
Ni	Nickel	179 ± 0.00	125 ± 0.03	273.00 ± 0.00
P	Phosphorus	261.40 ± 0.00	175.57 ± 0.06	275 ± 0.00
Pb	Lead	56 ± 0.01	67 ± 0.01	71.73 ± 0.02
S	Sulphur	127.00 ± 0.02	661.4 ± 0.02	868.20 ± 0.01
Zn	Zinc	8.8 ± 0.00	3.024 ± 0.00	10.08 ± 0.00

Data are average of mean ± SD

Table 9. Metal analysis from mixture of e-waste (video card and RAM) using ICP-MS

Symbol	Metals	Metal content (mg/g)		
		One step bioleaching	Two step bioleaching	Chemical leaching
Ag	Silver	0.001 ± 0.00	0.0010 ± 0.00	0.01 ± 0.00
Al	Aluminium	310 ± 0.03	325 ± 0.01	341.50 ± 0.02
Au	Gold	0.003 ± 0.00	0.022 ± 0.00	0.05 ± 0.00
Ca	Calcium	125 ± 0.01	398 ± 0.04	600 ± 0.00
Co	Cobalt	0.03 ± 0.00	0.3 ± 0.00	3 ± 0.00
Cd	Cadmium	17 ± 0.01	18 ± 0.00	20 ± 0.00
Cr	Chromium	0.094 ± 0.00	0.095 ± 0.00	0.8 ± 0.00
Cu	Copper	1252.77 ± 0.02	1293.16 ± 0.01	1300 ± 0.03
Fe	Iron	82.1 ± 0.01	87.8 ± 0.01	316 ± 0.02
K	Potassium	767.600 ± 0.05	873.600 ± 0.02	916 ± 0.00
Mg	Magnesium	301.700 ± 0.01	337.450 ± 0.00	345.64 ± 0.02
Mn	Manganese	3.23 ± 0.00	4.31 ± 0.00	4.79 ± 0.00
Mo	Molybdenum	0.007 ± 0.00	0.007 ± 0.00	0.7 ± 0.00
Na	Sodium	163.600 ± 0.01	149.700 ± 0.03	184.3 ± 0.03
Ni	Nickel	102 ± 0.01	240.62 ± 0.03	300 ± 0.04
P	Phosphorus	264.5 ± 0.02	300.6 ± 0.02	360 ± 0.00
Pb	Lead	45.1 ± 0.01	60.37 ± 0.00	75.37 ± 0.00
S	Sulphur	644.800 ± 0.02	852.800 ± 0.01	925 ± 0.00
Se	Selenium	0.075 ± 0.00	0.073 ± 0.00	10 ± 0.00
Si	Silicon	3.274 ± 0.00	3.686 ± 0.00	23 ± 0.00
Zn	Zinc	8.3 ± 0.00	9.7 ± 0.00	12 ± 0.00

Data are average of mean ± SD

CONCLUSION

Microorganisms possess the potential to thrive under hostile environmental conditions. To cope up with the adverse conditions, many bacteria have the ability to produce EPS. In the current study, attempt was made for the isolation and identification of EPS producing *Paenibacillus* sp. and its role in the leaching of metals from e-waste. *Paenibacillus* sp. was found to be excellent for leaching of metals viz., copper, cadmium, sodium and lead. Efficiency of *Paenibacillus* sp. for the extraction of metals varies with the source of e-waste. These results showed that chemoorganotrophic bioleaching can be substitute to chemolithotrophic bioleaching. Exopolymer layer is required for leaching of metals from e-waste. Colloidal chitin medium is found to be suitable for bioleaching of e-waste as it enhances the exopolymer layer of *Paenibacillus* sp. The bioleaching process by *Paenibacillus* will be fruitful to recover precious metals.

FUTURE PROSPECTS

- Further research should be carried out for extraction of precious and base metals using consortia of bacteria by bioleaching process.
- The bioleaching study should be carried on a large-scale.

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CONFLICT OF INTEREST

The authors declare that there is not any conflict of interests regarding the publication of this manuscript. In addition, the ethical issues, including plagiarism, informed consent, misconduct, data fabrication and/ or falsification, double publication and/or submission, and redundancy has been completely observed by the authors.

LIFE SCIENCE REPORTING

No life science threat was practiced in this research.

REFERENCES

Aneja, K. R. (2001). Experiments in microbiology, plant pathology, tissue culture and mushroom production technology. 3 rd edn. New Age International Ltd, New Delhi. 303-309.

Argumedo-Delira, R., Gomez-Martinez, M. and Soto, B. (2019). Gold bioleaching from printed circuit boards of mobile phones by *Aspergillus niger* in a culture without agitation and with glucose as a carbon source. Metals, 9; 1-10.

Batsala, M., Chandu, B., Sakala, B., Nama, S. and Sreenu, D. (2012). Inductively Coupled Plasma Mass Spectrometry (ICP-MS). Int. J. Res. Pharma. Chem., 2; 671-680.

Bellenberg, S., Leon-Morales, C., Sand, W. and Vera, M. (2012). Visualization of capsular polysaccharide induction in *Acidithiobacillus ferrooxidans*. Hydrometallurgy, 129-130; 82-89.

Buxton, R. (2011). Nitrate and nitrate reduction test protocols. Ame. Soc. Microbiol., 1-20.

Cornelissen, A., Ceyssens, P., T'Syen, J., Van Praet, H., Noben, J., Shaburova, O., Krylov, V., Volckaert G. and Lavigne, R. (2011). The T7-related *Pseudomonas putida* phage phi15 displays virion-associated biofilm degradation properties. PLoS One, 6; 1-11.

Cui, J. and Zhang, L. (2008). Metallurgical recovery of metals from electronic waste: A review. J. Hazard. Mater., 158; 228-256.

Darkoh, C., Chappell, C., Gonzales, C. and Okhuysen, P. (2015). A rapid and specific method for the detection of indole in complex biological samples. Appl. Env. Microbiol., 81; 8093-8097.

Govarthanan, M., Mythili, R., Selvankumar, T., Kamala-Kannan, S., Rajasekar, A. and Chang, Y. (2016). Bioremediation of heavy metals using an endophytic bacterium *Paenibacillus* sp. RM isolated from the roots of *Tridax procumbens*. 3 Biotech, 6; 242.

Haque, S. and Sao, S. (2015). Isolation and identification of microorganisms from different soil samples of Bilaspur (C.G). World J. Pharma. Res., 4; 2043-2057.

Ilyas, S., Munir, A., Niazi, S. and Ghauri, M. (2007). Bioleaching of metals from electronic scrap by moderately thermophilic acidophilic bacteria. Hydrometallurgy, 88; 180-188.

Kaliyaraj, D., Rajendran, M., Angamuthu, V., Antony, A, Kaari M., Thangavel, S, Venugopal G., Josep, J. and Manikkam, R. (2019). Bioleaching of heavy metals from printed circuit board (PCB) by *Streptomyces albidoflavus* TN10 isolated from insect nest. Bioresour. Bioproc., 6; 1-11.

Karastogianni, S., Girousi S. and Sotiropoulos, S. (2016). pH: Principles and measurement. In: Caballero, B., Finglas, P. and Toldra, F. (eds.) The Encyclopedia of Food and Health. 4; 333-338. Oxford: Academic Press.

Kavitha, A. (2014). Extraction of precious metals from e-waste. J. Chem. Pharm. Sci., 3; 147-149.

Kronenberg, P., Inaudi, D. and Smith, I. (2000). Development of an "optical hair"-hygrometer: A novel way to measure humidity using fibre optics. In: International conference on trends in optical nondestructive testing. Lugano, Switzerland. 1-8.

Kumar, C. (2014). Internet and e-waste awareness. Universal J. Environ. Res. Technol., 4; 227-234.

Li, J., Lu, H., Guo J., Xu, Z. and Zhou, Y. (2007). Recycle technology for recovering resources and products from waste printed circuit boards. Environ. Sci. Technol., 41; 1995-2000.

Li, X., Luo, K., Ren, J., Wang, X., Mu, Q. and Fan, W. (2017). Characterization of extracellular polymeric substances from different cyanobacterial species and their influence on biocalcification processes. Environ. Chem., 14; 254-265.

Madrigal-Arias, J., Argumedo-Delira, R., Alarcon, A., Mendoza-Lopez, R., Garcia-Barradas, O., Cruz-Sanchez, J., Ferrera-Cerrato, R. and Jimenez-Fernandez, M. (2015). Bioleaching of gold, copper and nickel from waste cellular phone PCBs and computer goldfinger motherboards by two *Aspergillus niger* strains. Braz. J. Microbiol., 46; 707-713.

Mitsunobu, S., Zhu, M., Takeichi, Y., Ohigashi, T., Suga, H., Jinno, M., Makita, H., Sakata, M., Ono, K., Mase, K. and Takahashi, Y. (2016). Direct detection of Fe (II) in extracellular polymeric substances (EPS) at the mineral-microbe interface in bacterial pyrite leaching. Microbes Environ., 31; 63-69.

Mrazikova, A., Marcincakova, R., Kadukova, J. and Velgosova, O. (2014). Nickel recovery from printed circuit boards using acidophilic bacteria. J. Polish Min. Eng. Soc., 2; 51-54.

Naumann, D. (2000). Infrared spectroscopy in Microbiology. In: Meyers R (ed) Encyclopedia of analytical Chemistry. John Wiley and Sons, Chichester, UK. 102-131.

Nocelli, N., Bogino, P., Banchio, E. and Giordano, W. (2016). Roles of extracellular polysaccharides and biofilm formation in heavy metal resistance of *Rhizobia*. Materials, 9; 1-19.

Pham. V. and Ting, Y. (2009). Gold bioleaching of electronic waste by cyanogenic bacteria and its enhancement with bio-oxidation. Adv. Mater. Res., 71; 661-664.

Pradhan, J. and Kumar, S. (2012). Metals bioleaching from electronic waste by *Chromobacterium violaceum* and *Pseudomonads* sp. Waste Manage. Res., 30; 1151-1159.

Rozas, E., Mendes, M., Nascimento, C., Espinosa, D., Oliveira, R., Oliveira, G. and Custodio, M. (2017). Bioleaching of electronic waste using bacteria isolated from the marine sponge *Hymeniacidon heliophila* (Porifera). J. Hazar. Mat., 329; 120-130.

Sajjad, W., Zheng, G., Din, G., Ma, X., Rafiq, M. and Xu, W. (2019). Metals extraction from sulfide ores with microorganisms: The bioleaching technology and recent developments. Trans. Ind. Inst. Met., 72; 559-579.

Tambekar, D., Hirulkar, N., Gulhane, S., Rajankar, P. and Deshmukh, S. (2007). Evaluation of hydrogen sulphide test for detection of fecal coliform contamination in drinking water from various sources. Afr. J. Biotech., 6; 713-717.

Willner, J. and Fornalczyk A. (2013). Extraction of metals from electronic waste by bacterial leaching. Environ. Protect. Eng., 39; 197-208.

Wu, W., Liu, X., Zhang, X., Zhu M. and Tan, W. (2018). Bioleaching of copper from waste printed circuit boards by bacteria-free cultural supernatant of iron-sulfur-oxidizing bacteria. Bioresour. Bioproc., 5; 1-13.

Xia, M., Bao, P., Zhang, S., Liu, A., Shen, L., Yu, R., Liu, Y., Chen, M., Li, J., Wu, X., Qiu, G. and Zeng, W. (2019). Extraction and characterization of extracellular polymeric substances from a mixed fungal culture during the adaptation process with waste printed circuit boards. Environ. Sci. Poll. Res., 26; 22137-22146.

Yazici, E and Deveci, H. (2013). Extraction of metals from waste printed circuit boards (WPCBs) in H_2SO_4- $CuSO_4$-NaCl solutions. Hydrometallurgy, 139; 30-38.