Process Biochemistry 46 (2011) 1589–1598

Contents lists available at ScienceDirect

Process Biochemistry

journal homepage: www.elsevier.com/locate/procbio

Bioleaching of flotation by-products of talc production permits the separation of nickel and cobalt from iron and arsenic

Kathryn D. Wakeman*, Petra Honkavirta, Jaakko A. Puhakka

Department of Chemistry and Bioengineering, Tampere University of Technology, PO Box 541, 33101 Tampere, Finland

ARTICLE INFO

Article history:
Received 3 December 2010
Received in revised form 28 April 2011
Accepted 29 April 2011

Keywords:
Arsenic
Bioleaching
Cobalt
Nickel
Iron
Flotation concentrate

ABSTRACT

Arsenic and iron can be found as impurities in nickel- and cobalt-containing flotation by-products of talc production. This study examined whether bioleaching could directly produce solutions containing nickel and cobalt but not iron and arsenic. In shake flasks, nickel and cobalt were over 99% dissolved from a nickel-rich flotation concentrate (NFC) and an arsenic rich flotation concentrate (GFC) around pH 3.0. During bioleaching of NFC (10% pulp density), arsenic remained below 20 mg/l and dissolved iron reached a maximum of 3.6 g/l. During bioleaching of GFC (10% pulp density), arsenic reached 4 g/l and dissolved iron remained below 20 mg/l. Decreasing the temperature from 27 to 6 °C reduced nickel and cobalt dissolution rates by factors of 13–24 and 14–24, respectively. In continuously stirred tank reactors (27 °C, pH 3.0), arsenic remained below 13 mg/l for NFC and reached 2.3 g/l for GFC. Dissolved iron was around 100 mg/l for NFC and below 5 mg/l for GFC. Bioleaching microorganisms enriched from the mine site included iron- and sulfur-oxidizing bacteria related to *Acidithiobacillus ferrooxidans*, *Acidithiobacillus ferrivorans*, *Acidithiobacillus caldus* and *Thiomonas cuprina*. These results show that nickel and cobalt can be recovered from NFC and GFC. Further, that dissolved arsenic can be controlled via biologically-mediated oxidation and subsequent precipitation of ferric iron.

1. Introduction

The term bioleaching refers to bacterial oxidation mechanisms that convert insoluble metal-containing minerals into a soluble form [1]. Acidophilic iron- and sulfur-oxidizing bacteria catalyze this process (e.g. [2–4]). Heap bioleaching is usually applied to low grade ores, whilst tank bioleaching is used for the treatment of ore concentrates. Continuously stirred tank reactors (CSTRs) are widely used for the bio-oxidative pre-treatment of concentrated gold ores [5,6] but only one commercial-scale bioleaching plant for the recovery of non-precious metals is currently in operation [7,8]. In industry, the use of CSTRs promotes high bioleaching rates and permits close process control.

Iron- and arsenic- containing sulfidic minerals (such as arsenopyrite; FeAsS) commonly occur in ores of precious and transition metals, and are present at elevated levels in ore concentrates. During CSTR-based processing, bio-oxidation of arsenopyrite results in the dissolution of arsenic and iron. Dissolved arsenic is toxic [9–11] and must be removed from leach liquors in a chemically stable state to prevent environmental contamination. Dissolved iron and arsenic also present deleterious metallurgical and process effects – by increasing the consumption of chemicals used for downstream processing of leach liquors and due to the toxicity of arsenic to many bioleaching microorganisms [12,13]. In the gold-mining industry, chemical treatments that are routinely used to remove arsenic and iron from leach liquors result in the formation of stable ferric arsenate precipitates (e.g. [14,15]). Biological removal of arsenic and iron from leach liquors can also result in the formation of stable iron and arsenic- containing precipitates [16–19].

Talc [$Mg_3Si_4O_{10}(OH)_2$] occurs as a secondary mineral in ultramific magnesite rocks, often in close association with (contaminant) transition metals. At a Finnish talc-mining site, froth flotation methods are used to remove nickel, cobalt, iron and arsenic from the crushed talc ore. The resultant sulfidic flotation concentrate contains valuable nickel and cobalt but also has a high iron and arsenic content. To reduce the arsenic level in the concentrate, gersdorffite [NiAsS] is removed by reverse flotation. This process results in significant losses of nickel in the form of pentlandite [$(Fe, Ni)_9S_8$]. In this work, the possibility of bioleaching these flotation side products of talc production for the recovery of nickel and cobalt was explored. The aim was to establish whether a solution containing high concentrations of nickel and cobalt and low concentrations of iron and arsenic could be directly produced by bioleaching. The

* Corresponding author. Tel.: +358 401981145; fax: +358 331152869.
E-mail address: kathryn.wakeman@tut.fi (K.D. Wakeman).

1359-5113/$ – see front matter © 2011 Elsevier Ltd. All rights reserved.
doi:10.1016/j.procbio.2011.04.016

diversity and composition of microbial communities enriched on each type of flotation concentrate were characterized using 16S rRNA profiling methods (PCR-DGGE and sequencing).

2. Materials and methods

2.1. Flotation concentrates

The flotation concentrates studied were (i) the product of the first flotation process, nickel flotation concentrate (NFC) and (ii) the reverse flotation product from NFC, gersdorffite flotation concentrate (GFC). The compositions of the flotation concentrate samples were as given in Table 1. For the determination of iron, nickel, cobalt and arsenic concentrations, flotation concentrates were fully dissolved in strong acid (65% HNO_3) and resultant solutions analyzed using acetylene flame AAS with Co, Fe, Ni and As lamps and the AAnalyst 400 (all from Perkin Elmer). Sulfur and carbon were analyzed from solid samples using an Eltra CS 500 carbon sulfur determinator (Eltra GmbH, Germany). Mineral compositions were approximated from elemental data. Acid consumption by NFC and GFC was similar in the bioleaching pH range (1.5–3.0; Table 2).

2.2. Media

Mineral salts medium (MSM) containing NFC or GFC at a pulp density (p.d.) of 1–10% was used to support the growth of bioleaching microorganisms. Final concentrations of mineral salts (stock solution pH = 1.8) and trace elements (stock solution pH = 1.5) in MSM were as follows: mineral salts (g/l de-ionized water): $(NH_4)_2SO_4$ (3.75); $Na_2SO_4{\cdot}10H_2O$ (1.875); KCl (0.125); K_2HPO_4 (0.0625); $MgSO_4{\cdot}7H_2O$ (0.625) and $Ca(NO_3)_2{\cdot}4H_2O$ (0.0175); trace elements (mg/l de-ionized water): $FeCl_3{\cdot}6H_2O$ (13.75); $CuSO_4{\cdot}5H_2O$ (0. 625); H_3BO_3 (2.5); $MnSO_4{\cdot}4H_2O$ (3.19); $Na_2MoO_4{\cdot}2H_2O$ (1.0); $CoCl_2{\cdot}6H_2O$ (0.75); $ZnSO_4{\cdot}7H_2O$ (1.125), and Na_2SeO_4 (1.125). Following addition of NFC or GFC, media pH was adjusted to that required using concentrated H_2SO_4 or NaOH. For shake flask studies, all media (containing NFC or GFC) were autoclaved (121 °C; 20 min) after pH adjustment. Media containing NFC or GFC were not sterilized prior to use in continuously stirred tank reactor experiments.

2.3. Monitoring of bioleaching

Bioleaching was monitored by analysis of liquor withdrawn from shake flasks or batch reactors. In all cases, any evaporation was redressed by the addition of sterile de-ionized water prior to sampling. Redox potential and pH were measured from non-filtered liquor within 120 min of sampling. Redox measurement employed a pH 340 redox meter (WTW, Germany) with BlueLine 31 Rx slimtrode electrode (Schott Instruments, Germany); pH was measured using a pH 315i pH meter with SenTix 81 slimtrode electrode (both from WTW, Germany). Liquor was filtered through 0.45 μm polysulfone membranes (Whatman, UK) prior to the following analyses: ferrous iron was measured using the ortho-phenantroline method, according to the American Public Health Association standard 3500-Fe [20] and a Shimadzu UV 1601 spectrophotometer. Dissolved total iron, nickel and cobalt were measured according to standards SFS 3044 and 3047 [21,22] using an atomic absorption spectrophotometer (AAS; Perkin Elmer 1100 B) fitted with Cu, Fe, Ni and Co lamps (all from Perkin Elmer Inc., Singapore). Arsenic was quantified using a graphite furnace AAS (AAnalyst 400) fitted with an As lamp (both from Perkin Elmer Inc., Singapore). Sulfate was analyzed by ion chromatography using a Dionex DX-120 fitted with an AS40 auto-sampler (Dionex Corporation, Sunnyvale, USA). Separation was achieved using an IonPac AS23 (4 mm × 250 mm) anion exchange column, preceded by an IonPac AG25 (4 mm × 50 mm) guard column. Samples were diluted in de-ionized water, and prepared according to standards SFS-EN ISO 10304-2 [23]. Eluent (0.5 M $NaHCO_3$/0.5 M Na_2CO_3 in de-ionized water) was degassed before use with under pressure; dissolution of gases to the eluent was prevented with helium overpressure during analysis.

Table 1
Percentage composition of flotation side products of talc production used in bioleaching studies.

Constituent	Flotation concentrate	
	Nickel	Gersdorffite
Total iron[Fe]	35.0	25.3
Sulfur [S]	30.7	36.7
Nickel [Ni]	10.5	14.3
Carbon [C]	2.1	0.2
Arsenic [As]	1.3	15.4
Cobalt [Co]	0.5	2.7
Nickeline [NiAs]	0.3	3.4
Gersdorffite[NiAsS]	2.5	30.6
Pyrrhotite[FeS)	36.5	20.2
Pentlandite[$(Fe, Ni)_9S_8$]	29.1	19.3
Pyrite [FeS_2]	8.4	12.1
Silicates/Carbonates	23.2	14.4

Table 2
Acid consumption of flotation side products of talc production in the pH range of 1.5–3.0. Acid consumption was determined at 1% pulp density. NFC: nickel flotation concentrate; GFC: gersdorffite flotation concentrate.

Final pH	Acid consumption ($g\,H_2SO_4\,kg^{-1}$)					
	Immediate		+4 h		+24 h	
	NFC	GFC	NFC	GFC	NFC	GFC
3.0	7	8	12	17	32	27
2.5	23	40	41	52	50	56
2.0	69	53	125	91	125	91
1.5	242	187	308	308	308	308

2.4. Preparation of inoculae for bioleaching experiments

To enrich indigenous microorganisms, sediment and water samples were obtained from a tailings pond (pH 4.6, redox 257 mV, 4 °C) at a Finnish talc mining site. All samples were stored at 4 °C and inoculated into MSM containing NFC or GFC at 1% p.d. within 24 h. Concomitantly, an enrichment culture (IS1) maintained on iron and sulfur and known to contain acidophilic iron and sulfur oxidizing microorganisms [24] was inoculated (10%, v/v) into media containing NFC or GFC at 1% p.d. All cultures were incubated at 27 °C (shaking, 150 rpm) for 24 days. The effect of inoculum origin on metals dissolution was studied during this enrichment/adaptation phase via weekly monitoring. After 24 days, enrichment cultures adapted to NFC were combined to form the mine-site NFC enrichment culture ('MS-NFC'); at the same time, enrichment cultures adapted to GFC were combined to form the mine-site GFC enrichment culture ('MS-GFC'). Both MS-NFC and MS-GFC enrichment cultures were maintained by regular sub-culturing into MSM containing NFC or GFC (pH 3.0; 1% p.d.), respectively. All maintenance/enrichment cultures were incubated (150 rpm) at 27 °C.

2.5. Shake flask bioleaching experiments

The effects of incubation temperature (6, 20 and 27 °C; 1% p.d.) and pulp density (2, 5 and 10% p.d.; 27 °C) on bioleaching of NFC and GFC were studied sequentially. All shake flask bioleaching experiments (250 ml shake flasks containing 100 ml media) were performed in triplicate at 150 rpm. The pH of shake flask liquors was adjusted to 3.0 prior to autoclaving and pH was not controlled during the incubation period. Active (3–4 day old) MS-NFC or MS-GFC sub-cultures were used to inoculate experimental systems (10%, v/v) containing NFC or GFC, respectively. Non-bioleaching (chemical) controls were prepared by addition of inoculum into shake flasks after pH adjustment and prior to autoclaving; contamination was prevented by the post-autoclave addition of 0.1% (v/v) of a solution of 5-methyl-2-(isopropyl)-phenol (2% w/v in methanol). Bioleaching parameters were monitored weekly.

2.6. Continuously stirred tank reactor experiments

Following completion of the shake flask studies, the effect of pulp density (2, 5 or 10% p.d.) on the bioleaching of NFC or GFC was tested in three 2 l stirred cylindrical glass batch reactors [25]. The use of CSTRs permitted pH control that was not possible in shake flasks. Each reactor contained 1.5 l of non-sterile MSM. After addition of NFC or GFC, the pH of the media was adjusted to 3.0. Reactors were inoculated (10%, v/v) with the relevant enrichment sub-culture and maintained at pH 3.0 by daily addition of acid/alkali, as required. The reactors were incubated at 27 °C, with stirring (160 rpm) and aeration (1.5 l of air min^{-1}) from a single inlet close to the base of the reactor.

2.7. Bacterial community composition and identification

Liquor (20 ml) from selected bioleaching experiments was passed through a 0.2 μm polysulfone membrane (Whatman, UK). Cells and sediment retained on the filter were washed by passing sterile de-ionized water (adjusted to pH 2.0 by addition of H_2SO_4) followed by sterile TRIS-buffer (pH 8.0; Sigma–Aldrich, UK) through the filter. Total DNA was extracted from filters using the Ultraclean Soil DNA Extraction Kit (MoBio Laboratories, USA) according to the manufacturer's instructions for maximum yield. PCR-amplified products were subjected to DGGE and unique bands were excised and sequenced, as described previously [26]. Identification of bacteria was via comparison of sequence data with that held in the National Center for Biotechnology Information (NCBI) database using MegaBLAST [27].