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## Geomicrobiology Journal

Publication details, including instructions for authors and subscription information:

<http://www.tandfonline.com/loi/ugmb20>

### Biohydrometallurgy for Nonsulfidic Minerals—A Review

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Version of record first published: 17 Aug 2010

To cite this article: Nalini Jain & D. K. Sharma (2004): Biohydrometallurgy for Nonsulfidic Minerals—A Review, Geomicrobiology Journal, 21:3, 135-144

To link to this article: <http://dx.doi.org/10.1080/01490450490275271>

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## Biohydrometallurgy for Nonsulfidic Minerals—A Review

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**Bioleaching is a technology applicable to metal extraction from low-grade ores, ore beneficiation, coal beneficiation, metal detoxification, and recovery of metals from waste materials. The technology is environmentally sound and it may lower operational cost and energy requirement. Whereas leaching of sulfidic minerals using chemolithoautotrophic bacteria is the most studied and commercially exploitable aspect of mineral biotechnology today, there is a dearth of literature on the dissolution of nonsulfidic minerals. Biohydrometallurgy of nonsulfidic minerals involves the action of heterotrophic microorganisms. Heterotrophic bacteria and fungi have the potential for producing acidic metabolites that are able to solubilize oxide, silicate, carbonate and hydroxide minerals by reduction, acidolysis and complexation mechanisms. It is an important aspect of biohydrometallurgy that requires development to meet future needs.**

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**Keywords** bioleaching, biohydrometallurgy, coal, metal recovery, demineralization

### INTRODUCTION

As civilization and technologies have advanced, the consumption of diverse mineral products has increased at a high rate. This has resulted in problems for metal production because many of the high-grade mineral deposits have become progressively depleted. As a consequence, metal production has to be met more often from lower-grade or complex ores, and from metal extraction from mining and industrial wastes (Torma 1986). Pyrometallurgical and hydrometallurgical methods may have some disadvantages such as poor product recovery, involvement of high process and energy cost and increase in pollution load of water resources. The recent upsurge of interest in biohydrometallurgical processes is motivated by the fact that they are relatively inexpensive, involving low energy

consumption and being environmentally safe. Biohydrometallurgy affords a simple and effective technology for extracting valuable metals like copper, gold, zinc, uranium, nickel, cobalt and many others from low-grade ores, rocks and waste materials by exploiting solubilizing and metal accumulating properties of microorganisms. Besides its industrial application to raw material supply, microbial leaching has some potential for remediation of mining sites, treatment of metal containing waste products and detoxification of sewage sludge (Bosecker 2001). It has become an advanced multidisciplinary technology involving chemistry, microbiology, and chemical and process engineering.

Bioleaching for metal extraction from sulfidic ores using chemolithoautotrophic bacteria such as *Thiobacillus ferrooxidans* (renamed as *Acidithiobacillus ferrooxidans* [Kelly and Wood 2000]) and *T. thiooxidans* is a well-known process and is being used commercially for the recovery of copper (Brierley and Brierley 2001; Pinches et al. 1997; Schnell 1997), gold (Aswegen 1993; Brierley 1994, 1997; Brierley et al. 1995; Olson 1994), uranium (Khalid et al. 1993; McCready and Gould 1990), cobalt (Briggs and Millard 1997; D'Hugues et al. 1999), nickel and zinc (Dew and Miller 1997) from their respective low-grade ores and high mineral concentrates. Microbial metal extraction from nonsulfidic minerals has received little attention to date.

Nonsulfidic ores such as oxides, carbonates and silicates contain no energy source for the microorganisms to utilize. Such ores may be leached by using heterotrophic bacteria and fungi, which require an organic carbon source as a source of energy and carbon for their growth. Bioleaching of nonsulfidic ores and minerals may be used for the recovery of valuable metals from low-grade ores and minerals as well as for the beneficiation of mineral raw materials, recovery of metals from wastes, and heavy metal detoxification of soils and solid residues.

The aim of this review is to provide an overview of heterotrophic leaching of nonsulfidic ores and minerals and to highlight the recent advances in the application of this process for metal extraction and detoxification of metal contamination. The effectiveness of these processes in extracting metals and avoiding pollution problems is discussed.

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Received 11 April 2002; accepted 6 October 2003.

Authors would like to thank the Council of Scientific and Industrial Research (CSIR), New Delhi, India for the financial assistance to carry out the research work.

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## MICROORGANISMS AND THE MECHANISM INVOLVED IN HETEROTROPHIC LEACHING

Almost all knowledge of biohydrometallurgy developed up to now deals with the use of chemolithoautotrophic bacteria for leaching of sulfidic minerals and ores. Leaching mechanisms of nonsulfidic minerals using heterotrophs have received less attention from microbiologists (Kiel and Schwartz 1980; Ralph 1985). Among the heterotrophic bacteria, members of the genus *Bacillus* and *Pseudomonas* have been found effective in the leaching of nonsulfidic minerals (Karavaiko et al. 1988). Fungi from the genera *Penicillium* and *Aspergillus* have also been used in mineral leaching (Ehrlich and Rossi 1990; Rezza et al. 2001).

Heterotrophic microorganisms require organic carbon as a source of energy and carbon. They produce metabolic by-products from the organic carbon they consume for energy production that may interact with a mineral surface. In addition to forming several organic acids such as acetic, citric, oxalic, and keto-gluconic acid (Agatzini and Tzeferis 1997; Castro et al. 2000; Natarajan and Deo 2001) (Table 1), heterotrophic microorganisms also produce exopolysaccharides (Malinovskaya et al. 1990; Welch and Vandevivere 1995; Welch et al. 1999), amino acids and proteins that can solubilize the metals via a variety of mechanisms. However, organic acids occupy a central

position in the overall process supplying both protons and a metal complexing organic acid anion (Gadd 1999)

**Bioreduction.** Some microorganisms can solubilize minerals by reduction. Such minerals include limonite, goethite, or hematite (Ehrlich 1986; Ferris et al. 1989). Ghiorse (1988) proposed that production of oxalic acid by a fungus can effect the reduction, of Fe (III) to Fe(II) thus increasing iron solubility. Bennet et al. (1997) observed a relationship between microbial colonization, iron reduction, and silicate weathering. They explained the reductive dissolution of iron oxide minerals and linked the silicate mineral weathering to microbial iron reduction primarily through the production of extracellular ligands. Hoffman et al. (1989) developed a process for the biological reduction of iron ore with *Pseudomonas* sp. Rusin (1992) and Rusin et al. (1994) suggested a process for bioremediation of heavy metal contaminated soil using an iron-reducing *Bacillus* strain.

**Acidification.** Lowering the pH to less than 5 results in an increased dissolution rate of many silicate and aluminium silicate minerals (Welch and Ullman 1996). Acidification may result either from the formation of an acidic metabolite or from a preferential utilization of alkaline substrate. Microbial oxidation of organic compounds may produce noncomplexing or weak complexing acids (carbonic, nitric, sulfuric, formic, acetic,

**Table 1**  
Microorganisms producing different organic acids during fermentation

Microorganism	Organic acid	Reference
<b>Bacteria</b>		
<i>Arthrobacter</i> sp.	Formic acid, acetic acid, oxalic acid, malonic acid, citric acid, phthalic acid	Liermann et al. (2000)
<i>Bacillus megaterium</i>	Citric acid	Krebs et al. (1997)
<i>Paenibacillus polymyxa</i>	Oxalic acid, acetic acid	Deo and Natarajan, (1997)
<i>Pseudomonas putida</i>	Citric acid, gluconic acid	Krebs et al. (1997)
<b>Fungi</b>		
<i>Alternaria</i> sp.	Citric acid, oxalic acid	Kovalenko and Malakhova (1990)
<i>Aspergillus</i> sp.	Oxalic acid, citric acid	Tzeferis (1994)
<i>A. niger</i>	Oxalic acid, citric acid, gluconic acid	Agatzini and Tzeferis (1997), Bosshard et al. (1996), Matty (1992), Meixner et al. (1985), Rezza et al. (2001), Schrickx et al. (1995), Singh et al. (2001), Strasser et al. (1994)
<i>Coriolus versicolor</i>	Oxalic acid	Sayer et al. (1999)
<i>Fusarium</i> sp.	Oxalic acid, malic acid, pyruvic acid, oxaloacetic acid	Bosecker (1989)
<i>Mucor racemosus</i>	Citric acid, succinic acid	Muller and Forster (1964)
<i>Penicillium</i> sp.	Citric acid, oxalic acid, itaconic acid	Agatzini and Tzeferis (1997), Parks et al. (1990)
<i>P. funiculosum</i>	Citric acid	Bosecker (1989)
<i>P. simplicissimum</i>	Citric acid, oxalic acid, gluconic acid	Borovec (1990), Burgstaller et al. (1992, 1994), Franz et al. (1991, 1993), Schinner and Burgstaller (1989), Tarasova et al. (1993)
<i>Streptomyces</i> sp.	Formic acid, acetic acid, oxalic acid, malonic acid, citric acid, phthalic acid	Liermann et al. (2000)

butyric, lactic, succinic, gluconic acid, etc.). Among the organic acids, 2-ketogluconic acid produced by some bacteria and citric acid and oxalic acid produced by some fungi have been shown to be very effective in the dissolution of silicates (Drever and Stillings 1997; Duff et al. 1963; Vandevivere et al. 1994; Welch and Ullman 1993). They furnish protons that help in breaking Si-O and Al-O bonds through protonation and catalysis.

**Ligand production/complexolysis.** Complexolysis is a process that utilizes microbially formed complexing and chelating agents that mobilize mineral constituents (Fe, Al, Cu, Zn, Ni, Mn, Ca, Mg, etc.) (Beveridge 1989). Microbes can produce and excrete organic ligands by a variety of processes such as fermentation and degradation of organic macromolecules (Berthelin 1983; Gadd 1999; Gottschalk 1986; Paris et al. 1996; Tzeferis and Agatzini 1994; Welch and Ullman 1999). These ligands can increase the rates of mineral weathering by forming stable soluble metal-organic complexes in solution, thereby increasing the solubility of the mineral (Amerhein and Surez 1988; Bennett et al. 1988; Wieland et al. 1988). In addition to low-molecular weight compounds, microbes also produce high-molecular weight compounds, such as microbial extracellular polysaccharides, that can enhance mineral dissolution by complexing with ions in solution, or they can inhibit dissolution by irreversibly binding to reactive sites on the mineral surface (Welch and Ullman 1999; Welch and Vandevivere 1995; Welch et al. 1999). A further mechanism of metal solubilization is the production of low molecular weight iron-chelating siderophores that specifically solubilize Fe (III) (Crichton 1991; Liermann et al. 2000). It has been reported by Bennett et al. (1997) that siderophores produced by bacteria to mobilize iron for cellular metabolism, also chelate and mobilize other metals. They showed that tropolonates and dihydroxybenzoates were quite reactive toward aluminum and silica, and both accelerated silicate dissolution. Over the past few years many siderophore or siderophore-like compounds have been identified from various biological systems. However the process of natural siderophore production by microorganisms needs to be optimized for application on an industrial scale.

**Alkalinization.** Biosolubilization of silicates is also possible via alkalinization of the media. The silicon-oxygen bond is disrupted under this condition. Avakyan (1985) demonstrated the release of silicon from nepheline, plagioclase or quartz by utilizing the bacteria *Sarcina ureae*. *S. ureae* grows in the presence of urea and produces ammonia resulting in the high alkalization of the medium.

## FACTORS AFFECTING BIOLEACHING PROCESS

Bioleaching processes need to be optimized with regard to the rate of bioleaching reactions, and with regard to the rate of growth of the microorganism involved. In order to optimize a leaching process, it is necessary to understand the nature of the biotic and abiotic reactions of the system. Factors that influence these reactions include the microbial population characteristics

as well as various physicochemical parameters and the properties of the material to be leached (Brandl 2001).

**Characteristics of the microorganisms.** Diverse microbes are capable of heterotrophic leaching. The process is affected by the size of the microbial population, its metal tolerance and adaptation abilities to the mineral environment. A variety of bacteria as well as fungi are known to be able to mobilize metals from nonsulfidic ores and minerals. However, they differ in their mode of action. It has been reported that the extraction of a specific element from a mineral depends to a great extent on microbial groups. Karavaiko et al. (1980) suggested that the mechanism of spodumene degradation for different microbial groups was related to the different metabolic products released into the medium by each group. These investigators found that fungi and thionic bacteria leached lithium and aluminium more actively in an acidic medium, whereas silicate bacteria leached silicon more actively in a weakly basic medium. Valix et al. (2001b) achieved enhanced nickel extraction with *Aspergillus niger* and better cobalt extraction with *Penicillium funiculosum* from low-grade laterite ores. Inoculum density has been found to affect the bioleaching rate of minerals. Burgstaller et al. (1992) found 50% enhancement in the rate of zinc extraction from filter dust on doubling the size of the inoculum (from  $2 \times 10^6$  to  $4 \times 10^6$  spores/ml of the medium) of *P. simplicissimum*.

In cases of leaching of metals from ores and waste, the toxicity of certain metals may influence the rate of leaching. The use of metal tolerant species enhances the leaching process (Bosecker 1985; Gadd and White 1989; Tzeferis et al. 1994; Valix et al. 2001a). Avakyan (1994) suggested that high concentrations of heavy metals act as a general protoplasmic poison, including denaturation of proteins and nucleic acid. He demonstrated an ability of microorganisms to survive in a heavy metal environment by selecting mutants at high concentrations of heavy metals. Isolation of nickel tolerant strains (Bosecker 1985; Tzeferis 1994; Tzeferis et al. 1994), nickel and cobalt-tolerant strains (Valix et al. 2001a) and aluminium tolerant strains (Kawai et al. 2000) have been successful. Valix et al. (2001a) observed that repeated subculturing of *P. simplicissimum* and *Aspergillus foetidus* in petri dishes with increasing metal concentration in the medium resulted in the appearance of mutants and adaptation of fungi within eight days, and the strains showed enhanced growth even at the high concentration (2,000 ppm) of heavy metals. They stated that the acclimatized strains acquired significant tolerance for heavy metals that could be leached from laterite ores and exhibited a potential for leaching the laterite ores.

**Physicochemical parameters.** Factors such as temperature, pH, oxygen supply, stirring rate, and nutritional composition of the medium have a direct influence on the leaching efficiency of microbes. Organic acid production by heterotrophs can be manipulated by changes in culture conditions (Xu et al. 1989; Schrickx et al. 1995; Dixon-Hardy et al. 1998) resulting in enhanced metal solubilization. Welch and Ullman (1999) studied the effect of temperature and pH on microbial metabolite production and mineral dissolution. They found 20-fold increase

of microbial dissolution of feldspar by decreasing the temperature down to 5°C causing an accumulation of gluconic acid in the medium that resulted in a significant reduction in pH. Vasani et al. (2001) reported that the ability of a metabolite produced by *Paenibacillus polymyxa* to dissolve calcium from low-grade bauxite ore decreased as the pH rose to near neutral. Rezza et al. (2001) observed the effect of length of incubation on dissolution of metals from an aluminosilicate (95% of spodumene) by heterotrophic microorganisms. They found that the amount of dissolved aluminum as well as the concentration of oxalic acid and citric acid in the medium fell after 15 days when leaching with *A. niger*. They found the same effect of length of incubation in leaching of the same mineral with *Rhodotorula rubra*. On leaching the aluminosilicate with *Penicillium purpurogenum*, a further enhancement in citric acid production and in the dissolution rate was observed even after 15 days of incubation. Oxygen supply has also been found to be an important parameter in the production of citric acid, which affects the leaching rate of zinc from filter dust by *P. simplicissimum* (Burgstaller et al. 1992; Franz et al. 1991). Burgstaller et al. (1992) observed that the interruption of the oxygen supply stopped citric acid production immediately. However, citric acid formation started again when oxygen was resupplied. Franz et al. (1991) showed that the total amount of citric acid excreted by the fungus increased from 101.8 mM to 136.8 mM on increasing the shaker speed from 150 rpm to 350 rpm, thereby increasing the oxygen supply. They also found an increased solubilization of zinc from industrial filter dust with increased shaker speed. The availability of carbon source and its concentration in the medium is found to be crucial for the growth of the heterotrophic organisms and metabolite production (Burgstaller et al. 1992; Castro et al. 2000). Alternative cheap and readily available carbon sources have been used (Groudev and Groudeva 1986; Strasser et al. 1994; Veglio 1996; Veglio et al. 1994) to optimize the cost of the leaching of metals on industrial scale.

*Properties of the material to be leached.* Pulp density of solid to be leached, particle size, mineralogical composition, effect of pretreatment, surface area and hydrophobicity of the solids are major factors in determining the rate and extent of any leaching. Burgstaller et al. (1992) reported that an increase in concentration of the leaching substrate such as filter dust from metal processing industries resulted in total inhibition of growth of *P. simplicissimum*. Vasani et al. (2001) observed an enhancement in the rate of Ca leaching at a pulp density of bauxite 5% as compared to 10%. Modak et al. (1999) reported that the leaching of a coarser particle of bauxite require more time than that of a finer particle. Khanna (1997) observed that the processing of bauxite of abrasive grade material might require the particle size to be -200+300 mesh. Vasani et al. (2001) also selected the -200+300 mesh (53–74 μm) size fraction of raw bauxite for leaching, suggesting the grinding of bauxite to finer sizes as an inevitable step for its biobeneficiation. Groudev and Groudeva (1986) found that prior activation of clays by heating at

600°C for 1 to 2 hours markedly increased their susceptibility to leaching.

## APPLICATIONS OF BIOLEACHING PROCESSES

It is possible to extract metals from nonsulfidic ores and industrial residues by applying bioleaching processes using heterotrophic microorganisms (Table 2). Bioleaching can also be used to remove unwanted metal impurities from ores (biobeneficiation). In addition it is possible to detoxify soil, sediment, and waste material polluted with heavy metals via bioleaching.

*Extraction of metals from low-grade ores and mineral concentrates.* Due to the uneven geographical distribution and rapid depletion of most of the conventional commercial quality ores and reserves, researchers are exploring the possibility of extracting and recovering valuable, precious and strategic metals from non-conventional resources, which are secondary raw materials and relatively cheap. Microbial leaching is a simple and effective technology used for metal extraction from low-grade ores and mineral concentrates (Krebs et al. 1997; Brandl 2001).

Recovery of metals from sulfide minerals using chemolithotrophic bacteria is the most studied aspect of biohydrometallurgy (Brierley and Brierley 2001). However, several reports exist, in which metals are extracted from silicate, carbonate and oxide ores through heterotrophic leaching (Groudev 1987; Burgstaller and Schinner 1993). Up to now, it has been demonstrated that heterotrophic microorganisms can increase the mobilization of elements (Si, Al, Fe, K, Li, Ni, Zn, and Mg) from rock forming minerals e.g., feldspar, pegmatite, hornblende and spodumene (Avakyan et al. 1981; Avakyan 1985; Barker et al. 1998; Kalinowski et al. 2000; Karaviko et al. 1980; Lierman et al. 2000; Rezza et al. 1997; Welch and Ullman 1999). Groudev and Groudeva (1986) used organic acid-producing heterotrophic bacteria and fungi for leaching of aluminium from clays and found the best results with a strain of *A. niger*. Manganese has been recovered from manganiferous minerals with the help of heterotrophic microorganisms (Ehrlich 1987; Toro et al. 1993; Veglio 1996; Veglio and Toro 1993; Veglio et al. 1993, 1995). Veglio et al. (1997) studied the technical feasibility of bioreduction of MnO<sub>2</sub> by heterotrophic mixed cultures on a pilot scale. They proposed an experimental flow sheet for the process, but the process was not cost effective. Ehrlich (2000) suggested the feasibility of biohydrometallurgical processes for the extraction of base metals such as Mn, Cu, Ni, and Co from ocean manganese nodules. Castro et al. (2000) investigated the role of bacteria from the genera *Bacillus* and *Pseudomonas* and of fungi from the genera *Aspergillus* and *Penicillium* in the leaching of zinc and nickel from two different silicate ores, calamine and garnierite. Bosecker (1985) and Tzeferis et al. (1991) showed the amenability of low-grade nickel oxide ores to leaching by *Aspergillus* sp. and *Penicillium* sp. Nickel recoveries as high as 90% have been observed (Sukla and Panchanadikar 1993). Agatzini and Tzeferis (1997) leached up to 60% and 50% of Ni and Co,

**Table 2**  
Heterotrophic microbes applicable to bioleaching processes

Organism	Substrate	Leached metal	Reference
<b>Bacteria</b>			
<i>Bacillus</i> sp.	Manganiferous ore	Ag	Rusin (1992)
	Electronic waste	Cu, Pb, Sn	Hahn et al. (1993)
<i>Paenibacillus polymyxa</i>	Calcite, hematite, corundum	Fe, Al, Ca	Deo and Natarajan (1997)
<i>Pseudomonas putida</i>	Filter dust	Zn	Muller et al. (1995)
	Fly ash from municipal waste incineration	Cd, Cu, Zn	Krebs et al. (1997)
<i>Arthrobacter</i> sp., <i>Nocardia</i> sp., <i>Pseudomonas</i> sp.	Spodumene	Li, Al, Si	Karavaiko et al. (1980)
<b>Fungi</b>			
<i>Aspergillus</i> sp.	Manganese ore	Mn	Ghiorse (1988)
	Laterite ore	Ni, Co, Mn	Tzeferis et al. (1994)
		Ni, Co, Fe	Valix et al. (2001b)
<i>A. clavatus</i>	Mercury compounds	Hg	Puerner and Siegel (1976)
<i>A. niger</i>	Copper ore	Cu	Dev and Natarajan (1981), Hartmann and Kuhr (1974), Kiel and Schwartz (1980)
	Manganese nodule	Cu, Ni	Ehrlich (1980)
	Coal fly ash	Al	Singer et al. (1982), Torma and Singh (1993)
	Clay	Al	Groudev and Groudeva (1986)
	Nepheline	Al	King and Dudeney (1987)
	$\beta$ -spodumene	Li	Ilger and Torma (1989)
		Li, Al	Rezza et al. (2001)
	Copper converter slag	Cu, Ni, Co	Sukla et al. (1992)
	Fly ash from municipal waste incineration	Heavy metal	Bosshard et al. (1996), Krebs et al. (1997)
	Laterite ore	Ni, Co	Agatzini and Tzeferis (1997)
	Electronic waste	Cu, Sn, Al, Ni, Pb, Zn	Brandl et al. (1999)
<i>A. ochraceus</i>	Silicate ore	Zn, Ni	Castro et al. (2000)
	Rocks	U	Munier-Lamy and Berthelin (1987)
<i>Penicillium</i> sp.	Manganese ore	Mn	Ghiorse (1988)
	Gold dust	Au	Groudev and Groudeva (1988)
	Silver ore	Mn, Ag	Gupta and Ehrlich (1989)
	Iron ore	Fe	Hoffmann et al. (1989)
	Laterite ore	Ni, Co	Agatzini and Tzeferis (1997)
<i>P. funiculosum</i>	Rocks	U	Munier-Lamy and Berthelin (1987)
<i>P. notatum</i>	Pagmetite rock	Li, Si, Al, Fe	Avakyan et al. (1981)
<i>P. simplicissimum</i>	Rocks	Ti	Silverman and Munoz (1971)
	Basalt rock	Al	Mehta et al. (1978, 1979)
	Industrial waste	Zn	Franz et al. (1993), Schinner and Burgstaller (1989)
	Tannery waste	Cr	Burgstaller et al. (1991)
	Filter dust	Zn	Burgstaller et al. (1992)
	Red mud	Al	Vachon et al. (1994)
	Electronic waste	Cu, Sn, Al, Ni, Pb, Zn	Brandl et al. (1999)
<i>P. purpurogenum</i>	Spodumene	Al, Li	Rezza et al. (2001)
<i>P. variotii</i>	Lead zinc ore	Zn	Dev and Natarajan (1981)
<i>Trichoderma ligneruum</i>	Pagmetite ore	Li, Si, Al, Fe	Avakyan et al. (1981)
<i>Yarrowia lipolytica</i>	Used catalyst	Cu, Pb, Sn	Hahn et al. (1993)
<b>Yeast</b>			
<i>Candida</i> sp.	Gold dust	Au	Groudev and Groudeva (1988)
<i>Rhodotorula rubra</i>	Spodumene	Al, Li	Rezza et al. (2001)
<b>Mixed culture</b>			
<i>Agrobacter radiobacter</i> , <i>Sphilococcus</i> sp., <i>Candida</i> sp.	Manganiferous ore	Mn	Veglio et al. (1997)

respectively, from nonsulfidic nickel ores using *Aspergillus* and *Penicillium* sp. They showed the presence of citric, oxalic and other organic acids in the leach liquors, indicating their role in the bioleaching process. Valix et al. (2001b) extracted up to 36, 54, and 0.76% of Ni, Co, and Fe, respectively, from low-grade laterite ores.

**Beneficiation of ores and coals.** Biobeneficiation refers to the selective dissolution of undesirable mineral component(s) of ores by microbial action thereby improving the quality of the ore. While ore bioleaching is a well-established phenomenon, processes as of biobeneficiation of ores are still being developed. For example, low-grade bauxite (<50% Al), the main source of aluminium is used in the manufacture of alumina-based abrasives and refractories. However, the presence of excess Ca and Fe (more than 0.5% as CaO and 1% as Fe<sub>2</sub>O<sub>3</sub>), respectively in low-grade bauxite ores is undesirable for use as an abrasive and in refractory applications (Deo et al. 1999; Modak et al. 1999; Vasan et al. 2001). Anand et al. (1996) used *P. polymyxa* to remove Ca and Fe selectively from bauxite. Biobeneficiation has also been applied to low-grade bauxite for the removal of silica (Groudeva and Groudev 1983).

In several countries such as India and Russia, low-grade coals have high mineral content, which adds to its transportation costs, risks of environmental pollution and lowers its calorific value (Sharma 1994). The coals usually contain insoluble metal silicates or oxides. Biobeneficiation of such coals is possible with the help of heterotrophic microbes. Disposal of large amounts of fly ash produced on burning of such coals is a major problem that raises economic and environmental issues. Although extensive research has been done on removal of sulfidic minerals from most of the U.S. coals, little has been reported on biobeneficiation of high ash producing coals. One such study by Sharma and Wadhwa (1997) used a mixed culture obtained from a biogas plant, which was enriched under aerobic conditions. They developed a two-repeated-step bioleaching process that resulted in removal of more than 50% of the mineral matter from low-grade Indian coals. Further studies are currently underway by the authors of this review.

**Recovery of metals from waste materials.** The size of ore reserves is limited and they may be exhausted unless they are conserved. To lessen dependence on ore reserves, utilization of different industrial as well as municipal wastes are now being considered as sources of metals. Bioleaching is a promising technology for recovering valuable metals, e.g., Fe, Zn, Ni, Co, Cr, and Cu from waste materials. Such metals can be recovered and recycled by metal processing industries. Wastes contaminated with valuable metals may be considered to be secondary resources. Bioleaching of the valuable metals from coal fly ash may afford a viable and economical process in the future within appropriate economic constraints (Sabati and Fleminger 1994; Siedel and Zimmels 1998). Filter dust generated during pyrometallurgical processes is of great economic value because of the presence of metal oxides in it. The possibility of leaching of zinc from filter dust was demonstrated with the fungus *P. simplicissimum*

(Schinner and Burgstaller 1989; Franz et al. 1991; Burgstaller et al. 1992). Sukla et al. (1992) used *A. niger* for leaching of Cu, Ni, and Co from copper converter slag. Krebs et al. (1997) have used *Pseudomonas putida*, *Bacillus megaterium*, and *A. niger* for the mobilization of elements like Cd, Cu and Zn from fly ash obtained from municipal waste incineration. Brandl et al. (1999) found that fungal strains (*A. niger*, *P. simplicissimum*) were able to mobilize 65% Cu and Sn and more than 95% Al, Ni, Pb, and Zn in electrical and electronic waste materials after a prolonged adaptation time.

Bosecker (1986) studied the use of organic acids in the leaching of red mud, a waste produced in the alkaline extraction of alumina from bauxite. The author extracted ~47% aluminium using oxalic acid. Vachon et al. (1994) increased by 75% solubilization of Al from red mud at a pulp density of 10% (v/v), using the organic acids produced by *P. simplicissimum*.

It may be possible to remove and recover metals by bioleaching not only from wastes but also from some catalysts used in chemical processes.

**Heavy metal detoxification.** Large quantities of toxic metals are discharged into the environment from the ever-increasing production and manufacture of goods and services and from the burning of fossil fuels to generate energy. Metallic pollutants have slowly worked their way into the atmosphere and the furthest reaches of the globe and unavoidably into the human food chain. The presence of heavy metals in wastes is a major constraint on safe waste disposal and recycling. Concerns have been expressed regarding proper control of pollution caused by mercury emissions from the combustion of coal (Neil et al. 1999). Biobeneficiation is a technique that may be successfully applied in the solubilization of toxic heavy metals from various solids. Brandl (2001) and Krebs et al. (1997) have summarized research efforts on this problem. Bioleaching using heterotrophic microorganisms has also been studied in the remediation of solid wastes and soils at high pH (Groudev 1987; Burgstaller and Schinner 1993; Hahn et al. 1993; Krebs et al. 1997). Some research work on bioleaching and biotransformation of low-grade Indian coals is currently underway in the research laboratory of the authors of this review.

## PROS AND CONS OF USING HETEROTROPHS

Whereas leaching by heterotrophs to solubilize metals from solids is possible at high pH, metal leaching by most of the autotrophic bacteria is possible only at acidic pH (Groudev 1987; Burgstaller and Schinner 1993; Hahn et al. 1993; Krebs et al. 1997). Most non-sulfidic ores contain no energy source for the growth of chemolithoautotrophic bacteria, most of which depend on the oxidation of sulfur or reduced iron and sulfur compounds. Therefore, the use of heterotrophic microbes is the agents of choice in leaching most non-sulfidic ores and minerals.

The use of heterotrophs in metal extraction from non-sulphidic ore has not received much attention for several reasons. The more neutral pH range at which these organisms grow

allows for easier microbial contamination to occur. Process sterilization is costly and presents a technical problem for a large-scale operation. The need for an organic carbon source for the growth of the heterotrophs and the production of leaching agents by them is another cost consideration. If cheap organic wastes generated in agriculture, in the food industry or in biotechnological processes can be used as growth substrates, leaching with fungi may be economic on an industrial scale.

The use of fungi presents a problem, however. The material to be leached is often adsorbed to or enclosed by the fungal mycelium. This is especially undesirable in the beneficiation of coal, bauxite and other materials. In this case the process must either be performed in two separated steps: (1) metabolite production and (2) leaching by the metabolite (Groudev and Groudeva 1986; Vachon et al. 1994; Brandl et al. 1999), or it may be performed with help of a dialysis membrane that separates coal from the fungus (Rezza et al. 2001). However, the use of dialysis membrane may not be feasible on an industrial scale.

The leaching of minerals by heterotrophs, mostly by the organic acids such as citric acid, lactic acid, gluconic acid and succinic acid that they produce, could be of great commercial interest to pharmaceutical and specialty chemicals industries. This may afford a link-up of the fermentation processes with the bioleaching operations.

## FUTURE OUTLOOK

In the future, the mining industry is expected to turn increasingly to bioleaching as a more economic way to process low-grade ores and mineral spoils that might otherwise be abandoned. The leached and recovered metals can be recycled as raw materials in metal manufacturing and chemical production. By reducing the metal content of coal residues, environmentally friendly materials can be produced that can be used for differ-

ent applications such as additives for construction materials, as adsorbing agents, or as enhancers of soil fertility.

Bioleaching is not sufficiently developed to be profitable in the recovery of zinc, nickel, cobalt and other metals from low-grade ores. The application of bioleaching to the beneficiation of silicate, carbonate, and oxide ores has not yet led to industrial applications. Research in this area has been intermittent and only on a laboratory scale. If heterotrophic organisms are the only organisms that can be applied in a given process, the choice should fall on the one that can grow at the expense of the least complex and least costly nutrients. Some industrial wastes (organic, nutritional, food, agricultural, etc.) may be utilized as carbon source for the fermentation processes.

A major problem associated with bioleaching is that it is slower than conventional techniques. Genetic engineering of the leaching organism(s), improvement in microbial fermentation techniques, engineering design of fermenters and applying enzymatic techniques may overcome this problem.

Although the use of heterotrophic microorganisms for nonsulfidic ore leaching has been extensively investigated, biobeneficiation of coals having high mineral matter content containing mostly oxides and silicates has been less studied. The biobeneficiation of low-grade coals has economic potential for coal-utilizing power sectors in most of the developing Asian countries such as India, where, the increasingly high amount of ash produced is alarming from both an economic and an environmental point of view. Because the composition of typical fly ash from Indian coal is very similar to that of nonsulfidic ores previously studied in heterotrophic leaching (Table 3), heterotrophic leaching may be applicable to this process. We are involved in the studies on bioleaching of low-grade Indian coals to remove undesirable mineral constituents to obtain cleaner fuels. We are using heterotrophs for biobeneficiating high-ash producing Indian coals.

**Table 3**  
Mineral composition of a few geological materials

Oxide	Material (% by wt.)					
	Hornblende <sup>a</sup>	Chlorite <sup>a</sup>	Garnieritic <sup>b</sup>	Haematic <sup>b</sup>	Clay <sup>c</sup>	Coal fly ash <sup>d</sup>
SiO <sub>2</sub>	44.3	35.1	52.23	41.50	44.47	58.0
Al <sub>2</sub> O <sub>3</sub>	16.2	10.7	0.18	18.43	36.08	28.0
Fe <sub>2</sub> O <sub>3</sub>	11.1	ND	8.18	19.36	0.93	7.0
MgO	12.2	21.5	16.87	2.08	—	2.0
CaO	8.9	0.3	8.51	3.42	0.22	2.5
Na <sub>2</sub> O	2.8	0.04	—	—	0.16	1.6
K <sub>2</sub> O	0.5	0.01	—	—	0.36	1.6
TiO <sub>2</sub>	0.8	ND	—	—	0.89	1.5
P <sub>2</sub> O <sub>5</sub>	<0.1	ND	—	—	—	0.4

<sup>a</sup>Liermann et al. (2000).

<sup>b</sup>Agatzini and Tzeferis (1997).

<sup>c</sup>Ambikadevi and Lalithambika (2000).

<sup>d</sup>Banerjee et al. (2000).

ND = Not detected.

Coal may be subjected to biobeneficiation in coal piles. Considering the fact that coal is stored for days and months prior to use by thermal power stations and by other industries, a slow rate of bioleaching may not be a problem. It may be necessary to integrate physical, chemical, and biochemical processing of coal and other ores, wastes etc. The authors of this review are currently doing research in this area. This research may have to be extended to the treatment of wastewater obtained from the bioleaching plants. There appears to be a good opportunity for the recovery of metals from these wastes.

More interdisciplinary cooperation between microbiologists, chemists, metallurgists and engineers is needed to identify, characterize, select, and develop bioprocesses for industrial and commercial applications. It offers a major opportunity for further research work in this area. Heterotrophic leaching holds great promise for the development of extraction technology for non-sulfidic ores.

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