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REVIEW PAPER

Advances in microbial leaching processes for nickel extraction from lateritic minerals - A review

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Abstract−Lateritic nickel minerals constitute about 80% of nickel reserves in the world, but their contribution for nickel production is about 40%. The obstacles in extraction of nickel from lateritic minerals are attributed to their very complex mineralogy and low nickel content. Hence, the existing metallurgical techniques are not techno-economically feasible and environmentally sustainable for processing of such complex deposits. At this juncture, microbial mineral processing could be a benevolent approach for processing of lateritic minerals in favor of nickel extraction. The microbial mineral processing route offers many advantages over conventional metallurgical methods as the process is operated under ambient conditions and requires low energy input; thus these processes are relatively simple and environment friendly. Microbial processing of the lateritic deposits still needs improvement to make it industrially viable. Microorganisms play the pivotal role in mineral bio-processing as they catalyze the extraction of metals from minerals. So it is inevitable to explore the physiological and bio-molecular mechanisms involved in this microbe-mineral interaction. The present article offers comprehensive information about the advances in microbial processes for extraction of nickel from laterites.

Keywords: Nickel, Lateritic Nickel, Microbial Mineral Processing, Fungi, *Acidithiobacillus*

INTRODUCTION

Nickel constitutes about 3% of the earth's composition and is the 24th most abundant element found in the earth's crust. The extensive use of nickel in steel and alloy industries is attributed to its exceptional blend of properties like high ductility, mechanical strength, good conductivity of heat and electricity, catalytic and anti-corrosion properties. Hence, application of nickel is increasingly coupled with global infrastructure development. The nickel-bearing minerals found in nature are broadly classified as sulfidic and lateritic (oxidic) types. Sulfidic minerals are rich in nickel content and thus industrially exploited for extraction of nickel throughout the globe. On the other hand, lateritic (oxidic) minerals are hardly utilized because of their mineral complexities and having lower nickel percentage. With the rapid increase in nickel consumption tied to gradual depletion of sulfidic reserves of nickel from earth crust induces to the exploitation of the underutilized laterite deposits for nickel production. However, technology to produce nickel from the major nickel reservoir, *i.e.*, lateritic deposits, is inadequate.

Lateritic form of nickel deposits represents major resources for nickel production. The laterites are mineral-rich, weathered ultramafic rocks deposited in earth crust. Laterization is the natural weathering process of ultramafic rocks in the earth crust to generate lateritic minerals. A warm climate and abundant rainfall are the favorable environmental conditions for the genesis of laterites, and hence the lateritic deposits are widely found in tropical regions of the world (Caledonia, Australia, Cuba, Brazil, Colombia, Greece, Philippines, Indonesia, and Sukinda valley of India) of the world [1-3]. Lateritic minerals are residual materials of weathered ultramafic rocks. Laterites are mostly composed of iron, aluminium, titanium, and manganese oxides because these are the least soluble components of the rocks undergoing weathering by nature. Thus the oxides of iron, aluminium titanium, and manganese constitutes the major mineral phases in lateritic minerals, and other value metals like nickel, cobalt and copper are associated with these mineral phases. The nickel-bearing lateritic minerals do not contain distinct nickel phase; rather nickel is embedded or interlocked in the secondary oxides of iron and silicate minerals [4]. Therefore, nickel laterites are broadly classified into limonite and saprolite types based upon the chemical nature of the nickel-bearing host minerals [5,6]. The limonitic laterites are iron oxide dominated minerals; however, the saprolites are silicate-enriched lateritic minerals. The majority of the lateritic deposits available on the earth surface are limonitic [$(Fe,Ni)O(OH)\cdot nH_2O$] type. Many reports suggest that oxides of iron, namely goethite [FeO(OH)], constitute a major chemical component of limonitic minerals, with which nickel is closely associated [7-9]. Generally, the silicate-rich saprolite types of laterites are found beneath the limonite zone, containing 1.5-2.5% nickel [4]. Laterite often contains minor amounts of cobalt and chromium along with nickel. On the other hand, the most commonly found sulfidic mineral of nickel in the earth crust is nickel pentlandite [$(Fe,Ni)_9S_8$], in which the major fraction of nickel is present in the form of iron sulfur complexes. Overall, the sulfide ores of the nickel contribute about three-quarters of the global nickel production,

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and out of which the pentlandite accounts for nearly 90% of the nickel producing sulfide ores.

Nickel from laterites is not easily extractable because of the complex mineralogy, low nickel content and heterogeneous distribution of nickel in lateritic minerals. Thus, the mineral processing cost for nickel production from complex lateritic minerals is too exorbitant; hence, the vast lateritic deposits have not been extensively used for nickel production so far [10]. On the other hand the sulfide minerals existing on the earth's surface are diminishing at an alarming rate due to their ruthless exploitation [11]. Hence, further need for the metal requires deep drilling of the nickel-bearing minerals deposited beneath the earth crust. So it is expected that the production cost of nickel will be much more expensive in the near future.

Generally, higher grade lateritic minerals are processed industrially through pyro- and hydrometallurgical routes. In pyro-metallurgical method, the ores are subjected to process through certain energy-consuming processes like drying, calcination, roasting, reduction, and smelting, and in case of hydrometallurgical methods, acids and solvents are used for treatment of ores to leach out of metals. Industrially, the nickel extractions from the lateritic minerals are also done by a process called as "Caron process," in which the both pyro and hydrometallurgical procedures are followed for nickel extractions from lateritic minerals. These metallurgical processes are economically viable and industrially acceptable, when the extraction of nickel is done from the relatively higher grade lateritic minerals. However, the majority of lateritic minerals available in the globe are poor in nickel content, hence the existing pyro and hydrometallurgical procedures are more energy- and capital cost consuming procedures [7]. But looking to the need of the day, the underutilized lateritic nickel deposits are needed to process for nickel production. In this context, the scope of the microbial mineral processing technology has been gaining momentum in mineral processing.

Microbial processing of minerals is an interdisciplinary process which involves the application of microorganisms for extraction of metal values at ambient conditions. Application of microbes for mineral processing has been extensively studied for the recovery of copper, gold, cobalt and uranium metals [13-15]. Microbial processing offers many advantages over the conventional metallurgical mineral processing methods for recovery of metals from low-grade ores and minerals due to its simple operations, low energy and less capital investment, and eco-friendly nature [16,17]. Though microbial mineral processing has drawn much attention in recent decades, a thorough understanding of the underlying mechanisms is needed to improve the efficiency of these processes. In bio-processing of minerals, the microbes act as catalysts for leaching out metals from complex minerals or acting as pre-treatment agent to alter the mineral structure, thereby making the minerals susceptible to lixiviants or leaching agents.

The microorganisms which play the pivotal role in the bio-processing of minerals are broadly classified into heterotrophic and autotrophic according to their nutritional behavior. The heterotrophic microbes are non-photosynthetic; they derive energy for their metabolism from organic carbon sources. These microorganisms secrete metabolites like hydroxycarboxylic acids, exo-polysaccharides, and these microbial metabolites are metal complexing and chelating agents and help in metal dissolution by lowering the pH of the medium [18]. On the other hand, the autotrophic microorganisms are photosynthetic; they use atmospheric CO_2 as the carbon source. But, the chemolithotrophic group of microbes belongs to autotrophic microorganisms that rely upon reduced inorganic compounds of iron and sulfur for their nutrition. The reduced iron and sulfur compounds present in the sulfidic minerals can support the nutritional requirement of chemolithotrophic microbes. Hence, this group of microorganisms has been widely applied for the processing of sulfidic minerals. Several researchers have been reported on the use of different strains of fungi for the processing of lateritic minerals and chemolithotrophic microorganisms for processing of sulfidic minerals. The use of chemolithotrophic microorganism in processing of lateritic ore has been discouraged since the laterites are devoid of nutritional support (reduced iron, sulfur compounds) for them. However, the acidophilic chemolithotrophic microbes belonging to genera *Acidithiobacillus*, *Leptospirillum* etc., have been used successfully for metal extraction from sulfidic minerals in large-scale for the commercial extraction [19]. But, the nickel extraction from lateritic mineral through microbial bio-processing route at industrial scale has not been achieved so far. Nevertheless, looking to the importance of the metal it became inevitable for development of a sustainable microbial process for nickel recovery from laterites. With this introduction, the present review article is focusing upon the advancement in microbial processing of lateritic minerals for nickel extraction.

MICROBIAL PROCESSING OF LATERITIC MINERALS

1. Application of Heterotrophic Microbes in Processing of Laterites

Heterotrophic microorganisms belong to different strains of fungi, and bacteria are the most widely studied microorganisms in bio-processing of lateritic minerals. The fungal strains of *Aspergillus* and *Penicillium* and bacterial strains of *Bacillus* and *Pseudomonas* have been widely studied microbial strains for recovery of metal values [12,20-26]. However, the fungal strains of *Aspergillus* and *Penicillium* are the most prominent microbial strains used in bio-processing of lateritic minerals for recovery of nickel.

1-1. Metal Chelating Microbial Metabolites in Metal Extraction

Mostly, heterotrophic microbes are non-photosynthetic and depend upon heterotrophic mode of nutrition. These microbes synthesize organic acids organic acids (citric, oxalic, gluconic etc.), as metabolic by-products during their cellular metabolism. These metal chelating properties and the acidity generated by these organic metabolites are responsible for the solubilisation of metal from lateritic minerals [27]. Other than the organic acids, the other microbial metabolites such as amino acids, exo-polysaccharides produced by these microorganisms are also reported to be involved in bioleaching of the metals [2,17,21,28]. The acidity generated by the cellular metabolites (carboxylic acids) of fungi is involved in breakdown of the metal-oxygen bond of lateritic minerals. The proton ions of organic acids first attack the oxygen atoms in the metal-oxide compounds of laterites. Most studies suggest that the citric acid and oxalic acid, which are the major organic metabolites produced by

the heterotrophic fungi, have a pivotal role in metal bioleaching from oxidic minerals. Some authors suggested for citric acid produced by microbes are the most effective leaching agent; however, Sukla and Panchanadikar [29] and Tzeferis [21] found that nickel recovery from laterites are more by oxalic acid [21,29]. Sukla and Panchanadikar [29] suggest that the oxalic acid effectively solubilizes the iron oxides (goethite) mineral phase in the lateritic chromite overburdens and concurrently the nickel (associated with the goethite phase) solubilization was also more [29]. On the other hand, the possible explanation for least effective of oxalic acid might be attributed to precipitation of the leached nickel as nickel oxalate, which has very low solubility [21]. However, Tang and Valix [2] suggested that the extent of metal dissolution is dependent upon the acid activity (hydronium ion concentration) rather than the type of metabolic acids involved [2]. Subsequently, the protonated oxygen molecule is then hydrolyzed and consequently the metal present in oxide minerals is solubilized. The following equations are representing generalized reaction during acid hydrolysis of minerals during fungus mediated mineral bio-processing [28].

(i) Acid production by microorganisms:

$$\text{Glucose/Sucrose} \xrightarrow{Fungus} \text{Oxalic/Citric/Gluconic acid}$$

(ii) Metal dissolution by proton attack

$$NiO + 2H^{+} \longrightarrow Ni^{+2} + H_2O$$

During mineral bio-processing fungal cells consume the organic carbons in the culture medium and convert them into different types of organic acids like oxalic, citric, gluconic and fumaric acid. The biosynthesis of these acids by fungi involves cellular metabolic processes like glycolysis and tricarboxylic acid cycle (TCA cycle). For the citric acid synthesis the glucose is first converted to the pyruvate in glycolysis process. The pyruvate is then oxidized to carbon dioxide and water in the TCA cycle, and simultaneously it accumulates the citric acid. The biosynthesis of oxalate in fungi can be formed from the catalytic breakdown of C-C bond in oxaloacetate by oxaloacetate hydrolase enzyme (oxaloacetate acetylhydrolase, OAH) and from the oxidation of glyoxylate and glycolaldehyde [30,31]. However, the widely proposed cellular pathway for oxalate production is through the breakdown of acetoacetate by OAH enzyme [32-34]. The key enzyme of the process, i.e., oxaloacetate acetylhydrolase (OAH), is located in the cytoplasm of *Aspergillus niger* and catalyzes the conversion of oxaloacetate to oxalate and acetate [30]. The role of these organic acids in mineral bio-processing has been evaluated by different groups of researchers. Behera et al. [35] studied that the supplement of manganese to the culture medium of *Aspergillus niger* improved oxalate secretion by the fungi during bioleaching process, and as a result the nickel recovery from chromite overburden was improved [35]. The elevated oxalate secretion by the *Aspergillus niger* in response to addition of manganese was attributed to enhancement of the catalytic activity of oxaloacetate acetylhydrolase (OAH) enzyme present in *Aspergillus niger* cytoplasm. As earlier as mentioned, out of several proposed pathways, that oxaloacetate was generated from breakdown of pyruvate during glycolysis process, which later on was hydrolyzed to oxalate and acetate by cytoplasmic enzyme oxaloacetate acetylhydrolase (OAH). The key enzyme oxaloacetate acetylhydrolase (OAH) is located in the cytoplasm of *Aspergillus niger*, where it catalyzes the conversion of oxaloacetate to oxalate and acetate [30]. The OAH enzyme belongs to phosphoenol pyruvate mutase [PEPM/isocitratelyase (ICL)] family. The members of this super family act upon α-oxycarboxylate substrate and cleave C-C or P-C bonds. All the PEPM/ICL super-family members require divalent metal (manganese or magnesium) cofactors for their catalytic activities, where these cofactors play the role of mediators in the interaction between the enzyme and substrate [36].

$$\text{Oxaloacetate} + H_2O \xrightarrow{Oxaloacetate\ acetylhydrolase} \text{Oxalate} + \text{Acetate}$$

Oxalic acid has two carboxyl groups, so the possible complexes of nickel cations with oxalate anion are expressed as follows [12].

(i) $C_2H_2O_4 \leftrightarrow (C_2HO_4)^{1-} + H^{+}$ (pKa_1=1.27)

$(C_2HO_4)^{1-} + Ni^{2+} \leftrightarrow Ni(C_2HO_4)_2$

(*Nickel Oxalate Complex*)

(ii) $(C_2HO_4)^{1-} \leftrightarrow (C_2O_4)^{2-} + H^{+}$ (pKa_2=4.20)

$(C_2O_4)^{2-} + Ni^{2+} \leftrightarrow Ni\ (C_2O_4)$

(*Nickel Oxalate Complex*)

Similarly, nickel forms nickel citrate complexes with citric acid secreted by the fungi.

$C_6H_8O_7 \leftrightarrow (C_6H_5O_7)^{3-} + 3H^{+}$ (pKa_3=6.39)

$2(C_6H_5O_7)^{3-} + 3Ni^{2+} \leftrightarrow Ni_3\ (C_6H_5O_7)_2$

(*Nickel Citrate Complex*)

Similarly, the nickel forms complexes with other carboxylic acid formed during the process.

Behera and Sukla [37] studied the effect of synthetic surfactant Polyoxyethylene sorbitan monolaurate (Tween-20) on bioleaching of nickel from lateritic chromite overburdens by *Aspergillus niger* [37]. It was observed that during fungal bioleaching, the addition of surfactant to the fungal medium in low concentration favored nickel extraction from pre-treated lateritic chromite overburdens. The surfactant used in the medium favored higher rate of sucrose consumption by *Aspergillus niger* for its metabolism. In addition, the average size of fungal micelle generated in presence of surfactant was comparatively smaller than that of without surfactant. Thus the *Aspergillus niger* micelles of smaller size provided more surface area for microbe-mineral interaction. Hence, the microbial metabolites generated at mineral-microbe interface interacted more efficiently upon mineral matrix, so as a result the extraction of nickel was improved.

1-2. Metal Extraction by Bioaccumulation

Fungal cell accumulates the metal ion from the solution during the bioleaching process to maintain the equilibrium between solid and dissolved metals, which in turn favors the continuous solubilization of the metal [29]. Nickel bioaccumulation inside the *Aspergillus niger* cell was reported by Magyarosy et al. [38]. The study revealed that nickel was accumulated inside the cell when the *Aspergillus niger* was grown in liquid media containing nickel. Furthermore, the study confirmed that nickel was accumulated inside the cell in the form of nickel-oxalate complex. However, the nickel accumulation in *Aspergillus niger* cell was inhibited by protonophores like carbonyl cyanide *p*-(trifluoremetoxy) and phenyl hydrazone

(FCCP). These protonophores disrupt the proton gradient of the electron carriers in the electron transport chain; thus the metal uptake and accumulation is inhibited. It indicates that membrane transporters of the cell play prominent role in the metal uptake. Thus, the metal accumulation is a metabolically active phenomenon. The metal accumulation phenomenon offers heavy metals resistant property to the fungi, which is an added advantage for microbes used in metal bioleaching [2,28].

Microbial recovery of nickel from low grade nickel laterite ores has been studied by several authors in laboratory scale only. However, commercialization of the process has been less successful due to its longer processing period coupled with poor metal recovery [24]. However, several authors reported that microbe-assisted leaching is comparatively more efficient than chemical leaching since the microbiological activities are involved in metal leaching along with metal chelating organic acid when compared with chemical leaching by organic acids [2,4,17,21,28]. It was reported that physical attachment of the fungal cell onto mineral surfaces contributed high organic acid (formed at hyphal tips) concentration with adjacent to mineral surfaces without greatly affecting the pH of the whole medium [39].

The mineralogy of the ores has significant effect on the metal recovery during the leaching process. Majority of nickel is present in an absorbed state within the goethite matrix of the lateritic minerals. Due to low solubility and complex structure, the extraction of nickel from the goethite matrix is very difficult. However, it has been reported that due to thermal pre-treatment of lateritic ores, nickel recovery is enhanced through bioleaching [40-42]. Thermal pre-treatment alters the mineralogical constitution of the laterites which occurs by dehydroxylation of goethite matrix in the laterites [11,43,44]. Behera et al. [12] found that as a consequence of thermal pre-treatment, the laterites converted into a mesoporous-like structure, and the surface area of the laterite ore particle was also increased [12]. As a result of which it improved the interaction of microbial metabolites with the ore particle and increased the recovery of nickel.

2. Chemolithotrophic Microorganisms in Processing of Lateritic Minerals

The chemolithotrophic (autotrophic) microbes are the group of microbes which oxidize iron and sulfur compounds for their metabolism. Due to this nature of metabolism such microbes are widely used for bioleaching. The majority of chemolithotrophic microbes reported for mineral processing belong to genera of *Acidithiobacillus*, *Leptospirillum* and *Sulphobacillus* etc. [45,46]. Among these microbes, *Acidithiobacillus ferrooxidans* bacterium is more elaborately studied for bioleaching studies.

2-1. Microbial-oxidative Pathways in Mineral Processing

The oxidative bioleaching mechanism involves the production of Fe^{3+} ion from the bio-oxidation of Fe^{2+} by *Acidithiobacillus* bacterial strain [36]. For sulfidic mineral processing, chemolithotrophic bacteria belonging to the genus *Acidithiobacillus* involve oxidative bioleaching mechanism. Two mechanisms, namely thiosulfate mechanism and polysulfide mechanism, have been proposed for bioleaching of acid insoluble metal sulfides like pyrite (FeS_2) and molybdenite (MoS_2), and for acid soluble metal sulfides such as sphalerite (ZnS), chalcopyrite ($CuFeS_2$), respectively [47,48].

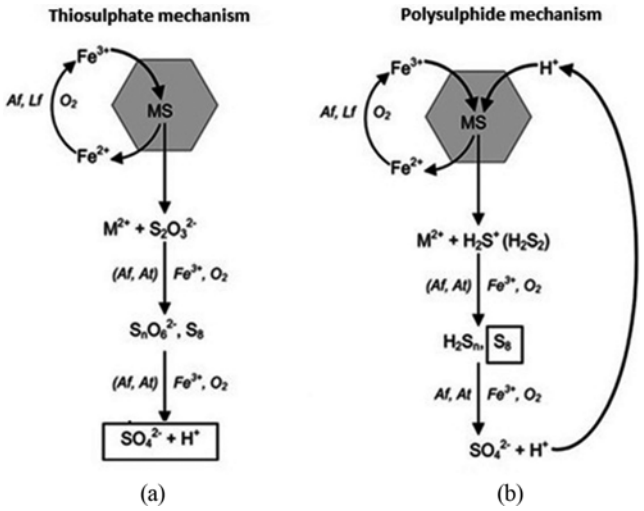

Fig. 1. Schematic comparison of the thiosulphate (a) and polysulphide (b) mechanisms in bio-leaching of sulphidic minerals [47]. Iron (Fe^{3+}) ions generated by the microorganisms attack upon metal sulphides (MS) and are subsequently reduced to the iron (Fe^{2+}) form. As a result, the metal sulphide bonds breakdown and releases metals as cations (M^{2+}).

2-1-1. Thiosulphate Pathway

In the thiosulfate mechanism, metal associated with sulfidic minerals is solubilized by ferric iron (generated by microbial process), which attacks the acid insoluble metal sulfides such as pyrite (FeS_2), molybdenite (MoS_2) or tungstenite (WS_2) and thiosulfate is generated as main intermediate product. Furthermore, the sulfate is generated as the main end product of the process [15]. The chemolithotrophic microbes belong to genera *Acidithiobacillus* and *Leptospirillum* convert ferrous iron to ferric form under aerobic condition. The active iron (III) acts as attacking reagent upon metal sulfide bond, and is subsequently reduced to the iron (II) form. As a result, the metal ions are released from metal sulfide crystals.The main role of the microorganisms in this mechanism is to catalyze the regeneration of the consumed ferric ions under aerobic condition as mentioned below. The schematic representation of this mechanism is presented in the Fig. 1(a), based upon the findings of the previous researchers [47,48].

2-1-2. Poly Sulfide Pathway

In case of the polysulfide mechanism, metal associated with sulfide minerals was solubilized by the combined attack of ferric iron and acid generated by the microbial activity. The metals associated with the acid-soluble metal sulfides such as sphalerite (ZnS), chalcopyrite (Cu_2S) or galena (PbS) are leached by this polysulfide process and element sulfur is the prime intermediate generated during this process. The elemental sulfur formed during the process is relatively stable, but can be oxidized to sulphate by sulfur-oxidizing microbes [15]. Thus, the role of microorganism in this mechanism is twofold: to catalyze the regeneration of the ferric ions consumed in the chemical oxidation of the intermediary hydrogen sulfide into elemental sulfur via formation of polysulfides. They catalyze the generation of sulfuric acid in order to maintain the supply of protons required in the first reaction step for the dissolution of the minerals. The schematic representation of this mechanism is pre-

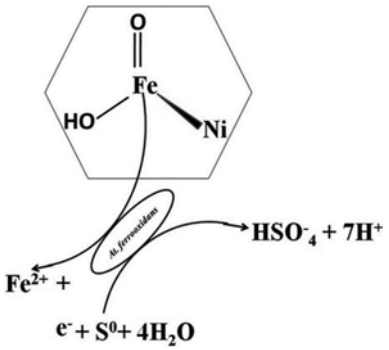

Fig. 2. Schematic presentation of anoxic microbial reduction process by *Acidithiobacillus ferrooxidans* for extraction of nickel from limonitic lateritic minerals [49,50,53,54]. Under anoxic condition *Acidithiobacillus ferrooxidans* reduces ferric iron (Fe^{3+}) of goethite [FeO(OH)] in limonitic lateritic minerals to ferrous (Fe^{2+}) form in presence of elemental sulphur (S^0) and subsequently elemental sulphur oxidised to sulphuric acid. The cumulative effect of reduction of ferric iron in goethite and production of sulphuric acid facilitates the dissolution of nickel present in association with goethite matrix of limonitic lateritic minerals.

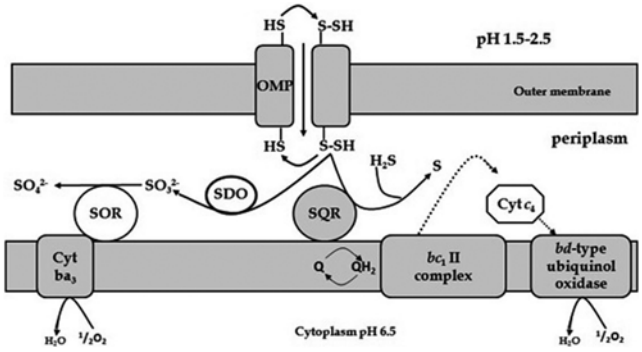

Fig. 3. A schematic presentation of biochemical model of elemental sulphur oxidation by *Acidithiobacillus ferrooxidans* cells based on findings of various researchers [54,66-68]. The thiol groups present in the outer membrane proteins of *Acidithiobacillus ferrooxidans* cells transport the elemental sulphur to the periplasm of the bacterial cell, where it is oxidized by a periplasmic sulphurdioxygenase (SDO) enzyme to sulphite and a sulphite acceptor oxidoreductase (SOR) to sulphate.

sented in the Fig. 1(b), as suggested by Schippers and Sand [47] and Rohwerder et al. [48].

Both the thiosulfate and polysulfide pathways suggest that a high microbial oxidation rate of ferrous to ferric iron is an important requirement for efficient bioleaching process of sulfide minerals. The acidophilic chemolithotrophic microbes rely upon reduced compounds of iron and sulfur for their metabolism. Hence these microbial strains have been successfully applied for bioleaching of sulfidic minerals. Since the laterites minerals lack in energy source for such chemolithotrophic microbes, hence the bio-oxidation process is hardly applied for the microbial processing of laterites.

2-2. Microbial Reduction Pathways in Mineral Processing

The microbial reductive method is a new development for processing of ferric rich lateritic minerals by using *Acidithiobacillus ferrooxidans* bacterium. This bacterium reduces ferric iron to ferrous iron in anoxic condition with a suitable electron donor (elemental sulfur) and produces sulfuric acid. Hallberg et al. [49] screened for the iron-reducing efficiency of *Acidithiobacillus ferrooxidans* under anoxic condition and reported its abilities to solubilize nickel from a limonitic laterite ore [49]. Behera et al. [50] reported for the successfully extraction of nickel from lateritic chromite overburden by using *Acidithiobacillus ferrooxidans* under anoxic environment [50]. In this process, Fe^{2+} ions were generated due to reduction of Fe^{3+} in the goethite during the microbial processing and the sulfur was oxidized to hydrogen sulfate (HSO_4^-) (which later on was converted to H_2SO_4) that generated acidity in the medium and was responsible for dissolution of nickel [51]. This can be explained by Eq. (1) as reported by Brock and Gustafson [52].

$$S^0 + 6Fe^{3+} + 4H_2O \longrightarrow HSO_4^- + 6Fe^{2+} + 7H^+ \quad (1)$$

The possible anoxic reduction of ferric iron by *Acidithiobacillus ferrooxidans* is due to the obligatory aerobic nature of the bacterium. Pronk et al. [53] demonstrated that *Acidithiobacillus ferrooxidans* is capable to growth on elemental sulfur in anaerobic condition, using ferric iron as an electron acceptor [53]. However, under aerobic condition oxygen behaves as a terminal electron acceptor during chemolithotrophic mode of respiration because ferrous iron in acidic environment is spontaneously oxidized to ferric iron and generates free electron.

The biochemical mechanism behind the sulfur metabolism by *Acidithiobacillus ferrooxidans* is schematically presented in Fig. 3. It involves the transport of elemental sulfur by the thiol groups present in the outer membrane proteins of the bacterium to the periplasm where it is oxidized by a periplasmic sulfur dioxygenase (SDO) to sulfite and a sulfite acceptor oxidoreductase (SOR) to sulphate [54].

Rawlings [54] lucidly reviewed the phenomenon; the Fe^{2+}/Fe^{3+} redox couple has a very positive standard electrode potential (+770 mV at pH 2) which is close to the standard electrode potential of O_2/H_2O redox couple (O_2/H_2O: +820 mV at pH 7); as a result, only oxygen bears the potentiality to act as a natural electron acceptor [54]. Hence under anoxic condition elemental sulfur behaves as the electron donor and ferric iron in goethite of laterite overburden acts as electron acceptor and is subsequently reduced to magnetite. The recent developments suggest that microbial processing of oxidic nickel ores might be technically feasible.

3. Dissimilatory Iron Reducing Bacteria (DIRB) in Lateritic Mineral Processing

Recentl, a number of bacteria have been isolated based on their ability to reduce several metal ions, including Fe (III) under anaerobic conditions. The popular iron-reducing microbial strains belong to the genera *Shewanella*, *Geobacter*, *Geovibrio*, etc. [55-58]. Dissimilatory iron reducing bacteria (DIRB) are a class of microorganisms which reduce Fe (III) coupled to the oxidation of organic compounds. IRB have been isolated and identified from broad environmental habitats [55]. DIRB utilizes Fe (III) oxides as terminal electron acceptors during bio-reduction process [59]. The biogeochemical cycle of iron is linked to metals co-associated with Fe (III) oxides or reduced iron phases (e.g., magnetite, siderite, or iron sul-

fide).

Various research groups suggested improved recovery of nickel through thermal pre-treatment of laterites [11,40]. This type of recovery is not cost effective in terms of energy consumption and may not be employed for large scale operations. Hence, it is desirable to have an alternative eco-friendly and low cost method, which makes the subsequent metal extraction more feasible. In this context microbial reduction caused by DIRB can be a possible alternative to thermal pre-treatment process in lateritic mineral processing. Microbial Fe (III) reduction results in the generation of several important Fe (II)-containing minerals in sedimentary environments, including magnetite $Fe(II)Fe(III)_2O_4$, which is a magnetic mineral. Magnetite formation during dissimilatory Fe (III) reduction is reported for different pure cultures of bacteria as well as for Fe (III)-reducing enrichment consortium [60,61].

Furthermore, Ester et al. [62] reported about the use of DIRB consortium in extraction of nickel from lateritic minerals [62]. The DIRB consortium acts as pre-treatment reagent for lateritic chromite overburden ore. The authors performed experiments of the bio-reduction of lateritic ore, and subsequently the DIRB treated ore was treated with H_2SO_4 for leaching of nickel. The nickel extraction from the DIRB pre-treated ore was about 68.5% by 8 M H_2SO_4 in 10 days, whereas the nickel recovery from untreated ore was 53.2% under similar conditions. The mineralogical analysis conducted by the authors revealed that the original lateritic ore contains goethite [(FeO(OH)] as major mineral phase along with minor phases of hematite, magnetite and quartz, and the majority of nickel associated with goethite phase similar findings were also reported before [6]. The DIRB treatment brought a characteristic mineralogical change in the ore sample. Goethite phase of the lateritic ore has been reduced to its dehydroxylated form of hematite or magnetite. Furthermore, the dehydroxylation of goethite is a complex phenomenon and it needs high thermal energy input for hematite formation. Thus, the use of a DIRB consortium offered the advantage of elimination of this energy-intensive thermal activation process. Subsequently, the DIRB treated ore was subjected to chemical leaching by H_2SO_4 showing encouraging outcome of nickel recovery. So, pre-treatment of laterites by DIRB brought about higher (15.3%) nickel recovery compared to the untreated laterites as reported by the authors. Very few studies have been conducted to compare the nickel extraction by using DIRB.

FUTURE PROSPECTS FOR MICROBIAL PROCESSING OF LATERITIC MINERALS

The heterotrophic microbes have been selected by several researchers for microbial leaching of lateritic ores. However, the use of heterotrophic microorganisms in extraction of metal from lateritic ores has been discouraging due to several drawbacks associated with the process. The fungal strains used in mineral processing show optimum growth at neutral pH range, which is more susceptible to contamination by other microbes. In this regard the maintenance of sterile environment for conducting such processes at larger scale is not economically sound. An added concern to the process is the need for organic carbon source utilized by the heterotrophs during the process. Furthermore, the use of fungi presents a setback in mineral processing because of undesirable production of excess fungal biomass. The fungal biomass and fungal mycelium often adsorb or accumulate the leached materials. Therefore, further recovery of metal from these fungal biomass and mycelium is also a matter of concern. Furthermore, cheap organic wastes generated from agriculture, domestic activities, food and beverage industries can be scientifically exploited as substrates for the growth of fungi.

The chemolithotrophic microorganisms (acidophilic, iron- or sulfur-oxidizing) have been studied more extensively in laboratory scale as well as in commercial scale [54]. The detailed molecular mechanism of bioleaching of sulfidic minerals by the model bacterial strain *Acidithiobacillus* has been studied elaborately and elucidated the detailed mechanism involved in microbial mineral processing. With the well-established phenomenon the strains of *Acidithiobacillus* bacteria have been used in industry scale operation for the recovery of copper and uranium [16,63-65]. In recent developments the *Acidithiobacillus* has been applied for the extraction of nickel from laterites in anoxic microbial reduction leaching process [49,50]. However, further detailed study of the molecular mechanism involving in the anoxic reduction process can be hoped for the application of chemolithotrophic microorganisms in the microbial processing of nickel laterites. During such a process, elemental sulfur was used for anoxic microbial reduction leaching of nickel from laterites; however, the process can be made economically sound by using low grade sulfide minerals in place of elemental sulfur. The application of low grade sulfide minerals instead of elemental sulfur can be regarded as a technology to generate wealth from waste.

DIRB has been used for nickel recovery at lab scale only. Very limited study has been conducted with DIRB. One of the most prospective technologies could be multistage microbial leaching of the laterites by using both DIRB and chemolithotrophic microorganisms.

CONCLUSIONS

Microbial mineral processing technology is a simple and effective technology for extraction of metal values from lean grade ores and mineral. This article focused upon the previous as well recent studies which were concerned with the extraction of nickel from lateritic ore through microbe-mineral interaction. Processing of sulfide minerals is based upon the activity of chemolithotrophic acidophilic microorganisms, mainly strains of *Acidithiobacillus* and *Leptospirillum* which convert insoluble metal sulfides into soluble metal sulfates. The lateritic or oxidic ores and minerals can be processed by heterotrophic microorganisms. In such cases, metal extraction is due to the production of organic acids and chelating compounds excreted by the microorganisms. The fungal mediated processing of laterites has been reported to have improved recovery of nickel, but still there remain some challenges in scaling up the process. On the other hand, the uses of the chemolithotrophic microorganisms for processing of lateritic (oxidic) minerals have been discouraged by conventional bio-oxidation leaching process, since the oxide minerals lack nutritional support (ferrous iron or reduced sulfur compounds) for such microbes. However, microbial reduction process by using DIRB and chemolithotrophic bacteria can provide a brighter avenue towards the development of a

techno-economically acceptable process of nickel extraction from laterites.

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