

Microbial Beneficiation of Iron Ore Collected from Rungta Mine Areas Using *Aspergillus fumigatus*

M. Pradhan 1 , M. Mishra 1 , C.C. Rath 2 *, L.B. Sukla 1

¹ CSIR-Institute of Minerals and Materials Technology, Bhubaneswar, India 2 Dept. of Botany, College of Basic Science & Humanities, Orissa University of Agriculture and Technology, Bhubaneswar, India

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Abstract: In the current study the fungus *Aspergillus fumigatus* was used to beneficiate the iron ore sample collected from Rungta mines, Odisha. The iron samples used in the experiment contains 58.2 % Fe, 5.7 % alumina and 5.0 % silica. Shake flask studies were carried out in Bromofield medium at 35°C, pH-6.8 and 150 rpm for 30 days. At the above conditions the maximum removal of alumina and silica by *A. fumigatus* was 24.5 % and 27.3 % respectively with drop in pH 1.8 from intial pH 6.8. Various optimization parameters like variation in growth medium,variation in pulp density and variation in temperature were studied with fungus. In Bromofield medium *A. fumigatus* observed to remove 24.5 % alumina and 27.3 % at optimum temperature 35 °C at 5 % pulp density. The results of the study indicated a potential relationship between Alumina and Silica removal and the organic acids production by this fungus. It is therefore concluded that there is a potential prospect in the use of metabolite from this type of fungus for biobeneficiation of iron ore minerals.

Key words: *Aspergillus fumigatus*, Alumina, Silica, Organic acid.

Introduction

Demand for iron and steel is increasing at a fast rate in the domestic market. The high grade iron ores are getting depleted, on the other hand, low grade ores and mining wastes are getting deposited at mining sites. High alumina content in the iron ore generates highly viscous slag hence it needs higher amount of fluxes and coke. Thereby producing huge volumes of slag as a result the productivity of the blast furnace is adversely affected. Increase in content of alumina by 1 %, increases the coke rate by 2.2 % which eventually decreases the productivity by 4 %. It also increases the consumption of flux by 30 kg/t of hot metal production 11 .

Now a days there is a greater interest in technologies that use microorganisms to mobilize

*Corresponding author (C.C. Rath)

E-mail: < chandicharanrath@yahoo.com > © 2014, Har Krishan Bhalla & Sons

or remove unwanted contaminants from valuable minerals. Such technologies, collectively referred to as biohydrometallurgy, are positively acknowledged for their environmental and economic advantages¹. Organic carbon is utilized by heterotrophic microorganisms as a source of energy and carbon. Organic acids are produced as by-products with utilization of this organic carbon source which interacts with the mineral surface. In addition to organic acids such as acetic, citric, oxalic, and keto-gluconic acid 2,5,15 heterotrophic microorganisms also produce exopolysaccharides ^{14,21}, 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

8 . The present work reports the potential application of fungus *Aspergillus fumigatus* for the bio beneficiation of low grade iron ore bearing high content of alumina and silica.

Materials and method *Mineralogical analysis*

Iron ore samples were obtained from Rungta mines, Odisha, India. The XRF study was carried out against the calibrated samples of similar values (Table 1). The size fraction of the iron ore used in these experiments is 75-60 microns.

Mineralogical analysis of the original and microbially treated ore was done using high resolution synchrotron based X- ray Diffractometer (XRD Philips model Diffractometer with CuK radiation).

Microorganism

Aspergillus fumigates ((GenBank Accession No: EF634391) was used for the beneficiation study.

Media

The medium used in this study was Bromfield medium [(g/L) sucrose-20, Yeast extract 1, K_2 HPO₄ 0.25, NH₄SO₄ 0.27, MgSO₄ 0.75, Sodium biphosphate 0.30, pH-6.8], CzapekDox broth $[(g/L)$ sucrose-30, $K_2 HPO_4$ 1, Kcl 0.5, NaNO₃3_, MgSO₄ 0.5, Ferrous sulphate 0.01, pH 7.3], Ashby [Glucose-20, Yeast extract 0.3, $K_2 HPO_4$ 0.2, $MgSO_4$ 0.2, Nacl 0.2, Kcl 0.1, $CaCo₃5.0$, pH-7.4 at 25^oc], MGYP medium[(g/ L) Glucose-10, Yeast extract 0.3, K_2HPO_4 2, $(NH_4)_2SO_4$ 1, KH_2PO_4 7.5, $MgSO_4$ 0.1, pH-6.8], Mineral salt medium[(g/L) sucrose-100, $\rm KH_{2}PO_{4}$ 1, $NH₄NO₃$ 3, $MgSO₄$ 0.5. All the media were procured from Hi-Media, Pvt. Ltd. Mumbai, India, prepared as per the manufactures' instructions and used in the study.

Shake flask beneficiation experiment

Experiments were carried out in 100 ml of Bromfield medium in 250 ml Erlenmeyer flask under sterile conditions at 150 rpm for 30 days. Each flask was inoculated by 10⁶ spores/ml of A. *fumigatus* and one flask was kept as control. 0.1 M HgCl_2 was added to the control flask to avoid contamination.

Effect of different growth media on biobeneficiation

The experiments were carried out at different growth medium such as Mineral salt, Bromofield ,Czapek, Ashby and MGYP. These media were inoculated with the cell culture of *Aspergillus fumigatus* to a concentration of approximately $10⁶$ spores/ml. The experiment was done at 10 % pulp density. The pH was maintained at 6.8 and the culture flasks along with the ore were sterilized in an autoclave for 20 minutes at 15 lb pressure to avoid any contamination. The flasks were then incubated at 35°C, 150 rpm for a period of 30 days. At the end of the experiment the samples were collected, dried and analyzed by wet chemical and XRF method.

Effect of temperature on biobeneficiation

The Rungta iron ore was subjected to beneficiation study at different temperatures such as 25, 30, 35 and 40. The experiments were carried out at an initial pH of 6.8 and 10 % pulp density for 30 days.

Effect of pulp density on biobeneficiation

Beneficiation studies were carried out at different pulp densities of iron ore such as 5, 10, 20 and 30 %. The pH of the medium was maintained at 6.8 and experiment was carried out at 35°C for 30 days. At the end of the experiment, solid residue was separated by filtration, dried in hot air oven and analyzed for Al, Si and Fe.

Results and discussion *Mineralogical analysis*

Hematite, magnetite, quartz and goethite were found to be the major iron bearing phases. The alumionousgaunge materials are present in form of kaolinite and gibbsite. The XRF analysis of the ore shows the presence of high alumina and silica content (Table 1).

Shake flask beneficiation study

Shake flask studies were carried out in bromofield medium at 35° C, pH-6.8and 150 rpm for 30 days. After the completion of the experiment, solid residue was filtered, dried and analyzed by XRF (Table 2). At the above conditions the maximum removal of alumina and silica by *A. fumigatus*

was 24.5 % and 27.3 % respectively (Figure 1). The initial pH of the medium was 6.8 which dropped down to 1.8 due to the production of organic acid during the study.

Effect of growth medium

Maximum removal of alumina and silica was achieved in bromofield medium. In this medium *A. fumigatus* observed to remove 24.5 % alumina

Constituents	Quantity in %
Fe ₂ O ₃	83.2
Fe (Total)	58.2
Al_2O_3	5.7
SiO ₂	5.0

Table 1. Chemical analysis of Rungta iron ore

Fig. 1. Alumina and silica removal by *A. fumigatus* in shake flask and the drop in pH at 10 % pulp density, 35° C and 150 rpm

and 27.3 % silica followed by MSM medium in which 23.45 % alumina and 26.33 % silica removal was obtained (Fig. 2 & 3). A wide range of media are used for isolation of different groups of fungi that influence the vegetative growth and

colony morphology, pigmentation and sporulation depending upon the composition of specific culture medium, pH,temperature, light, water availability and surrounding atmospheric gas mixture 13,16.

by *A. fumigatus* at 10 % pulp density, 35^oC and 150 rpm

Effect of temperature

30

Maximum removal of alumina and silica was observed at 35° C i.e. 24.5 % alumina and 27.3 % silica as it is the optimum temperature range for the growth of the fungus. At 30°C almost nearer percentage of removal was achieved i.e, 21.99 % alumina and 24.33 % silica (Fig. 4 & 5) in comparison to the control set. In the control set there is no removal of alumina or silica.

Effect of pulp density

The removal percentage of alumina and silica

Fig. 5. Effect of temperature on Silica removal by *A. fumigatus* at 10% pulp density and 150 rpm

decreased with increasing pulp density. The maximum removal was at 5 % pulp density. At this pulp density 30.98 % of alumina and 32.98 % of silica removal was achieved (Fig. 6 & 7). At 20 and 30 % pulp density the removal of alumina was 17.85 and 11.38 % respectively. The removal percentages of silica for the above pulp densities

were 19.22 % and 15.36 % respectively. This could be attributable to the availability of susceptible surface. The lesser the pulp density, more the sites are available for attack by the organic acids. In the control set there is no removal of alumina or silica.

The microorganisms convert the organic sugar

Fig. 6. Effect of pulp densities on Alumina removal by A. fumigatus at 35°C and 150 rpm

Fig. 7. Effect of pulp densities on Silica removal by *A. fumigatus* at 35°C and 150 rpm

source to different metabolites as organic acids and proteins. They attack on the mineral surface via different mechanisms. The organic acids have dual effect on the minerals. They lower the pH of the system hence dissolving the minerals and form complexes with the dissolved metals which lower their concentration in the solution thus allowing more solid to get dissolved ³. A wide variety of microorganisms have been reported to produce acidic metabolites, which are responsible in solubilization of alumino-silicates ¹². The production of organic acids has been established in both agro- and biomining industries as essential for natural dissolution of complex mineral materials by microorganisms ^{17,19}. Franz et.al.⁷ suggested that shaking of flasks during leaching is essential for production of organic acid and proper aeration.Microbes, basically fungus are well known for their ability to produce organic acids (oxalic, isocitric, succinic, malic, citric etc) and among them *Aspergillus* sp. has been most intensively 4,6,10. Biosynthesis of oxalic acid from glucose occurs by hydrolysis of oxaloacetate to oxalate and acetate catalyzed by cytosolic oxaloacetase. Citric acid is an intermediate of TCA cycle. Although the use of different microorganisms in ore leaching is well established, use of microorganisms in reducing alumina and silica from ore has been attempted in few investigations 15. Citric acid is a tricarboxylic acid and contains three carboxylic groups and one hydroxyl group as possible donor of protons (H^+) . When aluminum cations Al^{+3} are present in system and citric acid is fully dissociated in aqueous solution, a complexation reaction may take place 9,18:

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

Similarly, oxalic acid contains two carboxyl

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groups so the possible complexes of aluminium cation with oxalate anion are

 $C_2H_2O_4 \leftrightarrow (C_2HO_4)^{1} + H^+ (pKa_1 = 1.20)$ $3(C_2HO_4)^{1} + Al^{3+} \leftrightarrow Al(C_2HO_4)_3$ Aluminium oxalate complex or

 $C_2H_2O_4 \leftrightarrow (C_2O_4)^{1} + 2H^+ (pKa_1 = 4.20)$ $3(C_2O_4)^{1}$ + 2Al³⁺ \leftrightarrow Al₂(C₂O₄)₃ Aluminium oxalate complex

Conclusion

In the preset investigation, we have demonstrated the microbial beneficiation efficiency of *A. fumigatus* for removal of alumina and silica from low grade iron ore, collected from Rungta mines in Odisha. *A. fumigatus* could remove 24.5 % alumina and 27.3 % silica in 30 days in shake flask at 10 % pulp density, 35° C and 150 rpm is suggestive of its potential use in mineral beneficiation using microorganisms.After beneficiation the Fe(total) content increased to 60.65 % from original content 58.2 %. *A. Fumigatus* found to be involved in the leaching and beneficiation processes of silicate ores and minerals. The Si-O Si or Al-O frame work might be cleaved due to microbial action leading to the solubilization or the removal of cations from the crystal lattice of silicate causing the subsequent collapse of silicate lattice structure. Microbial beneficiation of low grade iron ore is a clean technology and may be better alternative to conventional methods to use the wastes generated from iron ore mining and processing.

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