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Effect of Particulate Sizes and Temperature on The Bioleaching rate of Iron from Silica-Sand withAspergillusniger and Penecillumnotatium.

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ABSTRACT: The effect of temperature and particle sizeson the bioleaching of iron from silica sand with two fungal strains of Aspergillusniger and Penecillumnotatiumhas been studied with three particle size fractions of silica sand: +106-212µm, +212-300µm, and +300-400µm over the temperature range 30 to 80^{0}C for seven hours. The experimental data show that the particulate sizes of silica sand affect both the amount of iron present in the sand and the amount of iron the microorganisms are able to leach. The smallest particle size fraction (+106 - 212) converted the most quantity of iron present in their sand fraction (27.189% and 25.784%) for A. niger and P. notatum respectively. Particle size +212-300 µm for both A. niger and P. notatum converted 24.686% and 23.809% respectively while +300-400µm converted 23.627% and 21.941% for A. niger and P. notatum respectively. the also show that temperature positively affect the ability of A. niger and P. notatum to leach iron from silica sand with 70°Cas the optimal temperature for bioleaching process for both Aspergillusniger and Penecilliumnotatum, since at this temperature, the maximum percentage conversion was achieved irrespective of the sand size fraction. When the temperature was increased to 80°C, the amount of iron leached fell, not minding the sand size fraction.

Keywords: Silica sand, Aspergillusniger, Penecillumnotatium, Particle sizes, Temperature.

I. INTRODUCTION

The growing world population has resulted in an increase in materials consumption and industrial expansion, causing unsustainable pressures on the planet's natural resource reserves and the environment. For this reason, investigation concerning the development of sustainable technologies and recycling strategies is becoming favoured (Monballiu, et. al., 2015). Mining and metallurgy have negative consequences for the environment (Maluckov B, et al 2017; Akcil and Koldas, 2006) and lead to changes in geological (Marschalko, et. al., 2010) and climatic conditions (Phillips J. 2016). Mining-metallurgy operations include the extraction, preparing of ores (crushing ore and flotation), processing of concentrates, and the disposal of waste rock and by-products. The main waste in mining and metallurgy are waste rocks, flotation tailings and slag, which often contain significant amounts of valuable metals; sometimes even more than the ore (Maluckov BS, 2017). Leaching is a critical operation in hydrometallurgy (FrankCrundwell2005).

Bacteria/Fungal leaching (bioleaching) process for extracting metals from their sulfide ores is a promising technology as an alternative to the traditional pyro-metallurgical process (Prabhu, et. al., 2019). As high-grade ore reserves are processed and in the absence of discoveries of new high-grade deposits, it is increasingly necessary to process ores of lower grade to meet demand. Mineral bioleaching is a method by which some low-grade ores can be processed economically. Metal extraction from low-grade sulfide ores and concentrates can be achieved with the assistance of acidophilic chemolithotrophic iron- and sulfur-oxidizing microorganisms (Bosecker, 1997; Watling, 2006).In the last decades, interest in bioleaching has increased significantly (Bosecker, K., 1997; Krebs, et al. 1997). Microbial leaching is more efficient compared to chemical routes because it is environmentally friendly: it neither uses nor releases dangerous chemicals, uses far less energy since it operates at room temperature, reduces initial and operational costs and maintains the crystal structure of clay to a large extent (Hajihoseini and Fakharpour, 2019).

In bioleaching, mineral-decomposing microorganisms are used to convert insoluble metal sulfides to soluble metal sulfates that can be readily recovered from the solution (Nkemnaso, et. al., 2018; Seifelnassr and Abouzeid, 2013; Christelet al., 2018). The solubilization of metals can be accomplished by various species of bacteria and fungi and is based on three main mechanisms that can occur simultaneously Acidolysis, complexolysis, redoxolysis (H. Brandl, 2001; Mishra, et al., 2005). In the mineral-decomposing process, AspergillusnigerandPenecillumnotatium and their consortium are considered as the most important fungal (Ndukwe and Chikwendu, 2010). The important microorganisms are iron-oxidizing Leptospirilliumferrooxidans and L. ferriphilum, sulfur-oxidizing Acidithiobacillusthiooxidans and A. caldus, and iron- and sulfur-oxidizing A. ferrooxidans(Deshpande, et. al., 2018)

Bioleaching is influenced by a wide range of parameters including physicochemical and microbiological factors. These affect both the growth of the microorganisms and their leaching behavior (Monballiu, et. al., 2015).Various researches have shown that microbial activities in dependent on various factors such as pH, temperature, particle sizes. Peyton et al., (2010) noted that lead, zinc, and copper sorption and the binding-site affinities of bioleaching microorganisms are highly dependent on pH and temperature; this agrees with Hajihoseini and Fakharpour, (2019) argument that the quantity of iron extracted by Aspergillusniger from kaolin is affected by temperature.Mustafa et. al., (2011) have shown that contact time, pH, incubation temperature and carbon source play important role in the bioleaching of iron from silica sand; this agrees with an earlier study by Ndukwe and Chikwendu (2010) and Ndukwe and Eze, (2017) who investigated the effect of temperature on the bioleaching of iron from silica-sand. They were able to show that temperature affects the ability of the microorganisms to bioleach.

For bioleaching to be successful, it is obvious that optimal growth conditions must be maintained and the microorganism must be able to leach the material. In addition, and most importantly, the microorganism must be resistant to the metals that are leached out (Lee, et. al., 2001).

Ndukwe and Eze, (2017) investigated the effect of particle size and temperature on the bioleaching of iron from silica-sand using one fungal strain but the present study used two strains (A. niger and P. notatium). The results were then compared with those for the initial sample.

II. MATERIALS AND METHOD..

Initial Concentration of Iron in the Silica Sand

2kg of naturally occurring silica sand was obtained from Unwana River in Afikpo North Local Government Area, Ebonyi State, Nigeria. The sample was washed with deionized water to remove clay particles and other unwanted materials adhering to the surface of the sand and dried for 3-days and separated into different particle size fractions with the aid of a set of manual sieves. Three different particle size fractions +106 - 212µm, +212µm - 300µm and +300 - 400µm each for Aspergillusniger and Penecilliumnotatium,were selected for the experiments.

The presence of iron in each of the particle size fractions were determined by the Thiocyanide solution method. Standard solutions containing 0.5 to 2.5mg/ml of Iron (III) were prepared and the absorbance of the solutions was determined with the aid of a UV/Visible spectrophotometer. The relationship between concentration of iron in the standard solution and absorbance was established with the measured data, and used subsequently to determine the concentration of iron in the Silica sand and in the leachate. 100g of silica sand from the first particle size fraction (+106 -212µm) was weighed and transferred into a 500ml conical flask containing 25ml of 8M HNO_3 (KERMEC) and made up to 250ml with distilled water. The mixture was agitated for about 5 minutes and subjected to digestion for about 60 minutes. The solution was cooled and then filtered into 250ml standard flask, after which 10ml of it was pipetted and its absorbance determined using the UV/Visible spectrophotometer at a wavelength of 630nm. This experiment was repeated for the particle size fraction +212µm - 300µm, and +300 -400µm respectively. The results are given in table 1.

Fungi Isolation, cultivation and Identification

The fungus was isolated from the soil at a refuse dump in Unwana, Ebonyi State, Nigeria. The culture Sabouraud Dextrose Agar (SDA) (TM, MEDIA, India) medium was prepared according to the manufacturer's instruction. 62g of the SDA was dissolved in 1000ml of distilled water and stirred thoroughly to ensure proper homogenization. It was allowed to soak for 10 minutes, mixed and sterilized by autoclaving at 121^{o}C for 15minutes and was cooled to 45^{0}C before pouring into sterile petri-dishes. 1g of the soil sample was weighed and dissolved in 9ml of sterilized formal saline. 0.1ml was pipetted out and spread evenly on solidified

SDA, and incubated at 30^0C for 10 days, so that adequate number of spores formed. The fungal isolates wereidentified based on their morphology and colour of the surface of the colony using fungal identification guidelines described by Samson et al (2001).

For microscopic identification, wet mounts of the isolates were prepared using lactophenol cotton blue. A drop of the lactophenol cotton blue was placed on a sterile grease-free slide with a sterile dissecting needle. A small portion of the inoculum was collected and teased on the slide. The preparation was covered with a cover slip and examined under a microscope using low (x10) and high power (x10) objective with the Irish sufficiently closed to give a good contrast, for the purpose of observing hypha cells and spores.

Bioleaching

2.0g of silica sand from the particle size fraction, +106 - 212µm, was weighed into 250ml conical flask containing 150ml of prepared medium which contained Sucrose (KERMEC) 100g/l; NH4NO3 (AVIS Chemicals 99%) 450mg/l; KH2PO4 (KERMEC) 100mg/l; MgSO4.7H2O (KERMEC) 300mg/l; FeSO4.7H2O (GFS Chemical) 0.1mg/l; ZnSO4 (KERMEC) 0.25mg/l as culture medium. The flask was plugged with a non-absorbent cotton and then sterilized in an autoclave (Searchtech YA-208A) for 20 minutes at 1210C. It was then inoculated with 2ml of Aspergillusnigerspore suspension (2 x 108 spores/ml) and agitated for 20 minutes to achieve a good degree of homogeneity between the fungi and the sand. The flask was incubated in an oven (DHG 9101-USA) at a temperature of 30^0C and monitored for 7 hours. At a regular interval of 1 hour, 10ml of the leachate from the flask was obtained, filtered, and digested to stop further microbial actions. The concentration of iron leached out was then determined with the UV/ Visible spectrophotometer, after which the fractional conversion of the iron was calculated. The experiment was repeated at the temperatures of 40°C, 50°C, 60°C, 70°C, and 80°C. A similar experiment was done with +212µm - 300µm, and +300 - 400µm sand size fractions for both fungi strains. The results are given in tables 2, 3 and 4 for Aspergillusniger; 5, 6, 7 for Penecillumnotatium respectively. This is according to Ndukwe and Chikwendu, (2010); O. C. N. Ndukweet al., (2017).

III. RESULTS AND DISCUSSION

Effects of Temperature

Table 1 show that particulate sizes of silica sand affect the Iron inherent in it. The highest amount of iron (48.045g/g sand) in the sand matrix was located in the smallest sand fraction (+106-212µm); the amount of iron identifiable decreased with increase in the size fraction. Since initial concentration of iron fell with increased sand size fraction, being 42.568mg/g sand for +212-300µm and 40.035mg/g sand for +300- 400µm, the effectiveness of Aspergillusniger and Penecilliumnotatumin the removal of iron from silica sand will be enhanced by beneficiating the sand. Figures 1 and 2 show that temperature positively affect the ability of A. niger and P. notatumto leachiron from silica sand. The quantity of iron leachate from the process increased as the temperature of the process increased regardless of the fraction size not the fungi strain present. Though the rate of bioleaching of iron is approximately the same with all the size fractions,Aspergillusnigerextracted more quantity of iron from the sand than Penecilliumnotatumfor all temperature and particulate size fractions with their maximums extractable percentage beaning 27.189% and 25.784% for A. niger and P notatiumrespectively. The tables 1, 2, 3, 4, 5, 6, and 7 and figures 1 and 2 show that 70^0C is the optimal temperature for bioleaching of iron from silica sand for bothAspergillusniger and Penecilliumnotatum, since at this temperature, the maximum percentage conversion was achieved irrespective of the sand size fraction. When the temperature was increased to 80^0C, the amount of iron leached fell, not minding the sand size fraction. This agrees with earlier research by Ndukwe and Eze (2017)

Effect of Particle Size

Figure 3shows that the particulate sizes of silica sand affect the amount of iron the microorganisms are able to leach. The smaller the particle sizes the more the microorganisms are able to leach the iron content of the sand for both A. niger and P. notatium. This is supported by Cammen (1982); Sessitschet. al., (2001). Thesmallest particle size fraction (+106 - 212)converted the most quantity of iron present in their sand fraction(27.189% and 25.784%) for A. niger and P. notatum respectively. Particle size +212-300µm for both A. nigerand P. notatum converted 24.686% and 23.809% respectively while +300-400µm converted 23.627% and 21.941% for A. nigerand P. notatum respectively

IV. CONCLUSION

The present study investigated the effect of particle size and temperature variations on the bioleaching of iron from silica-sand with two different fungi stains. It can be concluded from the study that particle size fractions as well as temperature variations have effects on not just the leached iron but on the initial iron content of the sand even when they are from the same source. It was also deduced that the optimal operating temperature for the bioprocess is 70°C. This was where the maximum quantity of iron leachate was gotten.

Table 1: Initial Concentrations of Iron in 100g Silica Sand of various Particle Sizes

PARTICLE SIZE (µm)	INITIAL CONC. OF IRON (mg/g sand)
+106-212	48.045
+212-300	42.568
+300-400	40.035

Table 2: Conc.- time data for Aspergillusniger, iron conc. and % of iron leached values with temperature for particle size of +106-212µm

	30°C			40°C			50°C			60°C			70°C			80°C		
Time (h)	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**
0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
1	0.008	1.578	3.284	0.012	2.478	5.155	0.016	3.378	7.031	0.020	4.279	8.906	0.025	5.405	11.250	0.024	5.180	10.782
2	0.012	2.477	5.156	0.016	3.378	7.031	0.020	4.279	8.906	0.025	5.405	11.250	0.031	6.757	14.064	0.028	6.081	12.657
3	0.017	3.604	7.501	0.021	4.505	9.377	0.025	5.405	11.250	0.030	6.732	14.012	0.034	7.432	15.469	0.032	6.982	14.401
4	0.023	4.954	9.562	0.027	5.856	12.189	0.033	7.207	15.001	0.036	7.883	16.408	0.041	9.009	18.751	0.037	8.108	16.876
5	0.032	6.919	14.401	0.036	7.883	16.408	0.040	8.784	18.283	0.043	9.459	19.688	0.046	10.135	21.095	0.044	9.685	20.158
6	0	7.8	16.	0.0	9.0	18.	0.0	9.9	20.	0.	11.	23.	0.0	11.	24.	0.0	11.	23

	.036	83	408	41	09	751	45	10	626	051	261	438	53	712	377	52	486	.907
7	0.040	8.784	18.283	0.047	10.360	21.563	0.051	11.261	23.438	0.055	12.162	25.314	0.059	13.063	27.189	0.057	12.613	26.252

Table 3: Conc.- time data for Aspergillusniger, iron conc. and % of iron leached values with temperature for particle size of +212-300µm

30°C				40°C			50°C			60°C			70°C			80°C		
Time (h)	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL(%)**	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL**
0	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.0000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
1	0.007	1.352	3.176	0.010	2.027	4.762	0.014	2.928	6.878	0.015	3.153	7.407	0.018	3.829	8.993	0.016	3.378	7.936
2	0.010	2.025	4.757	0.013	2.702	6.347	0.016	3.378	7.936	0.019	4.054	9.524	0.023	4.955	11.678	0.021	4.505	10.583
3	0.014	2.927	6.876	0.017	3.604	8.466	0.020	4.279	10.052	0.023	4.955	11.640	0.027	5.856	13.757	0.025	5.405	12.697
4	0.018	3.828	8.993	0.021	4.505	10.583	0.025	5.405	12.697	0.028	6.081	14.285	0.032	6.982	16.254	0.030	6.732	16.815
5	0.023	4.954	11.678	0.026	5.631	13.228	0.030	6.732	16.815	0.033	7.207	16.931	0.037	8.108	19.047	0.035	7.658	17.990
6	0.028	6.018	14.137	0.031	6.757	15.873	0.035	7.658	17.990	0.039	8.559	20.107	0.042	9.234	21.692	0.040	8.784	20.635
7	0.032	6.919	16.254	0.035	7.658	17.990	0.039	8.559	20.107	0.043	9.459	22.220	0.048	10.586	24.868	0.045	9.910	23.280

Table 4: Conc.- time data for Aspergillusniger, iron conc. and % of iron leached values with temperature for particle size of +300-400μm

30°C				40°C			50°C			60°C			70°C			80°C		
Time (h)	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**
0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
1	0.006	1.302	3.252	0.008	1.577	3.939	0.011	2.252	5.625	0.014	2.928	7.314	0.016	3.378	8.438	0.015	3.009	7.516
2	0.008	1.575	3.934	0.010	1.852	4.626	0.014	2.928	7.314	0.017	3.604	9.002	0.021	4.505	11.253	0.019	3.935	9.829
3	0.011	2.250	5.260	0.013	2.546	6.359	0.017	3.604	9.002	0.020	4.279	10.688	0.025	5.405	13.501	0.023	4.861	12.142
4	0.017	3.603	9.000	0.019	3.935	9.829	0.022	4.730	11.815	0.025	5.405	13.501	0.029	6.306	15.751	0.027	5.787	14.455
5	0.022	4.730	11.815	0.024	5.093	12.721	0.027	5.856	14.627	0.031	6.750	16.860	0.034	7.432	18.564	0.033	7.176	17.924
6	0.026	5.630	14.063	0.028	6.019	15.034	0.030	6.532	16.316	0.035	7.658	19.128	0.039	8.559	21.379	0.037	8.102	20.237
7	0.029	6.623	16.543	0.031	6.713	16.768	0.033	7.207	18.002	0.038	8.333	20.814	0.043	9.459	23.627	0.041	9.028	22.550

Table 5: Conc.- time data for Penecillumnotatium, iron conc. and % of iron leached values with temperature for particle size of +106-212µm

	30°C			40°C			50°C			60°C			70°C			80°C		
Time (h)	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL**	**LA**	**ICL**	**%ICL**
0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
1	0.006	1.302	2.710	0.010	2.025	4.215	0.014	2.927	6.092	0.018	3.378	7.031	0.023	4.953	10.309	0.022	4.730	9.845
2	0.010	2.025	4.215	0.014	2.993	6.230	0.018	3.828	7.968	0.023	4.953	10.309	0.029	6.623	13.785	0.026	5.631	11.720
3	0.015	3.153	6.563	0.019	3.935	8.190	0.023	4.954	10.311	0.028	6.018	12.526	0.032	6.982	14.532	0.029	6.623	13.785
4	0.020	4.278	8.906	0.025	5.405	11.250	0.030	6.732	14.012	0.034	7.432	15.469	0.039	8.558	17.812	0.034	7.432	15.469
5	0.029	6.623	13.785	0.034	7.432	15.469	0.037	8.102	19.047	0.040	8.784	18.283	0.043	9.459	19.688	0.041	9.028	18.791
6	0.033	7.207	15.001	0.039	8.558	17.812	0.042	9.234	19.219	0.048	10.585	22.031	0.050	11.092	23.087	0.048	10.586	22.034
7	0.037	8.102	19.047	0.044	9.685	20.158	0.048	10.586	22.034	0.052	11.487	23.909	0.056	12.388	25.784	0.053	11.712	24.377

Table 6: Conc.- time data for Penecillumnotatium, iron conc. and % of iron leached values with temperature for particle size of +212-300µm

30°C				40°C			50°C			60°C			70°C			80°C		
Time (h)	LA	ICL	% ICL	LA	ICL	% ICL	LA	ICL	% ICL	LA	ICL	% ICL	LA	ICL	% ICL	LA	ICL	% ICL
0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
1	0.005	1.026	2.410	0.008	1.578	3.707	0.012	2.478	5.821	0.013	2.702	6.347	0.016	3.378	7.936	0.014	2.927	6.876
2	0.008	1.578	3.707	0.011	2.252	5.290	0.014	2.927	6.876	0.017	3.604	8.466	0.021	4.505	10.583	0.019	3.828	7.710
3	0.012	2.478	5.821	0.015	3.153	7.407	0.018	3.828	8.993	0.021	4.505	10.583	0.025	5.405	12.697	0.023	4.954	11.638
4	0.016	3.378	7.936	0.019	3.828	8.993	0.023	4.954	11.638	0.026	5.631	13.228	0.030	6.532	15.815	0.028	6.018	14.137
5	0.021	4.505	10.583	0.025	5.405	12.697	0.028	6.018	14.137	0.031	6.757	15.873	0.035	7.658	17.990	0.033	7.207	16.931
6	0.026	5.631	13.228	0.030	6.732	15.815	0.033	7.207	16.931	0.037	8.102	19.033	0.040	8.784	20.635	0.038	8.333	19.576
7	0.030	6.723	15.794	0.034	7.432	17.459	0.037	8.102	19.033	0.041	9.009	21.164	0.046	10.135	23.809	0.042	9.244	21.716

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Table 7: Conc.- time data for Penecillumnotatium, iron conc. and % of iron leached values with temperature for particle size of +300-400µm

	30℃			40℃			50℃			60℃			70℃			80℃		
Time (h)	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**	**LA**	**ICL**	**% ICL**
0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
1	0.004	0.750	1.873	0.006	1.302	3.252	0.008	1.578	3.941	0.011	2.252	5.625	0.013	2.702	6.749	0.012	2.248	5.615
2	0.006	1.302	3.252	0.008	1.578	3.941	0.011	2.252	5.625	0.014	2.927	7.311	0.018	3.829	9.564	0.016	3.378	8.438
3	0.009	1.901	4.748	0.011	2.252	5.625	0.014	2.927	7.311	0.017	3.603	9.002	0.022	4.730	11.815	0.020	4.279	10.688
4	0.015	3.153	7.876	0.017	3.604	9.002	0.019	3.935	9.829	0.022	4.730	11.815	0.026	5.630	14.063	0.024	5.093	12.721
5	0.020	4.279	10.688	0.022	4.730	11.815	0.024	5.093	12.721	0.028	6.019	15.034	0.031	6.757	16.878	0.030	6.532	16.316
6	0.024	5.093	12.721	0.026	5.630	14.063	0.027	5.856	14.627	0.032	6.919	17.282	0.036	7.880	19.682	0.034	7.432	18.564
7	0.027	5.856	14.627	0.029	6.623	16.543	0.030	6.732	16.815	0.035	7.658	19.128	0.040	8.784	21.941	0.038	8.333	20.814

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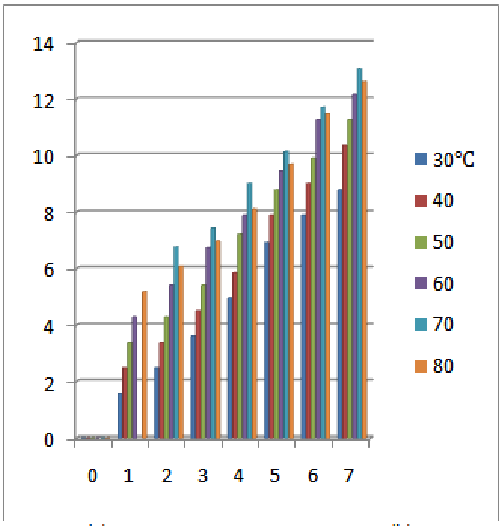
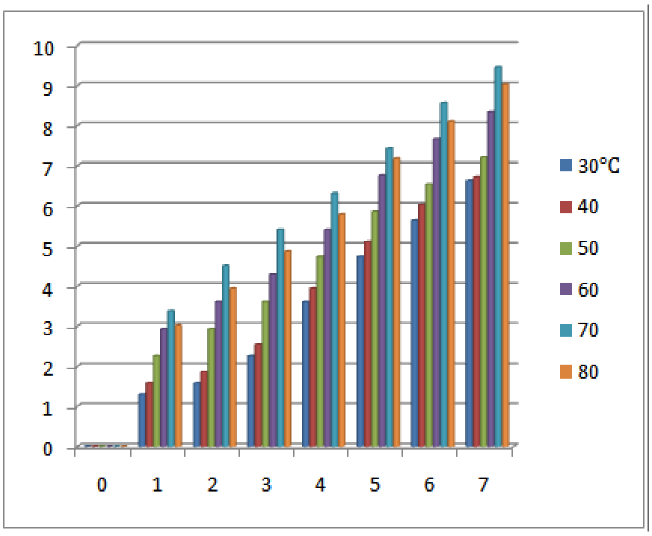


(a)



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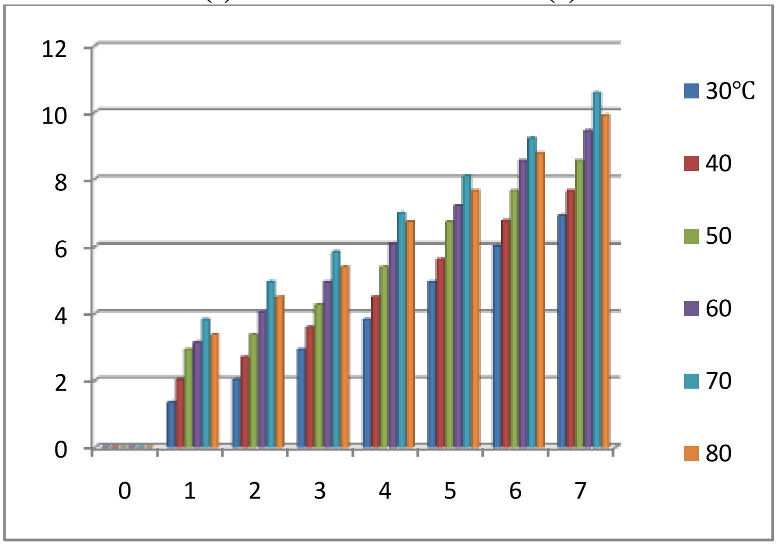

(c)

Figure 1: Conc.- time data for iron leached values by Aspergillusniger with temperature and different particle size fractions (a) represent particle size fraction of +106-212µm; (b) particle size fraction of +212-300µm; (c) represent size fraction of +300-400µm

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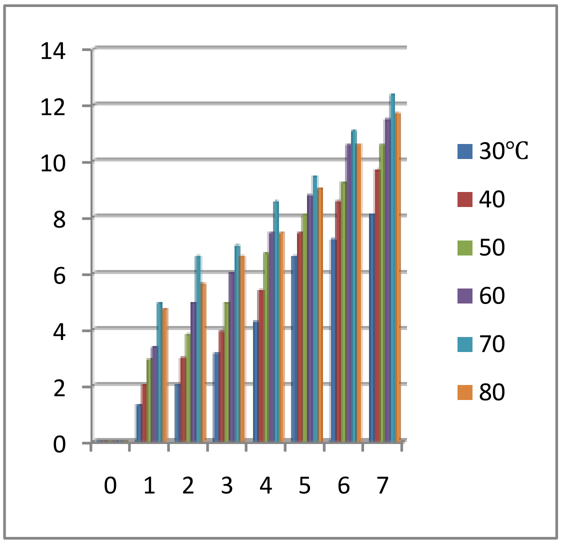

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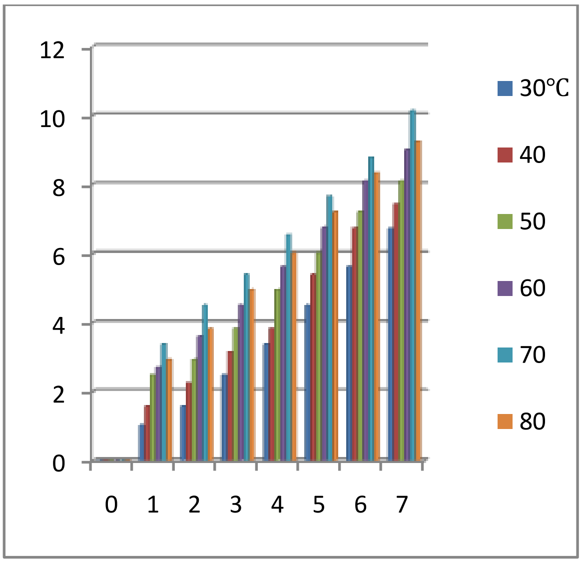

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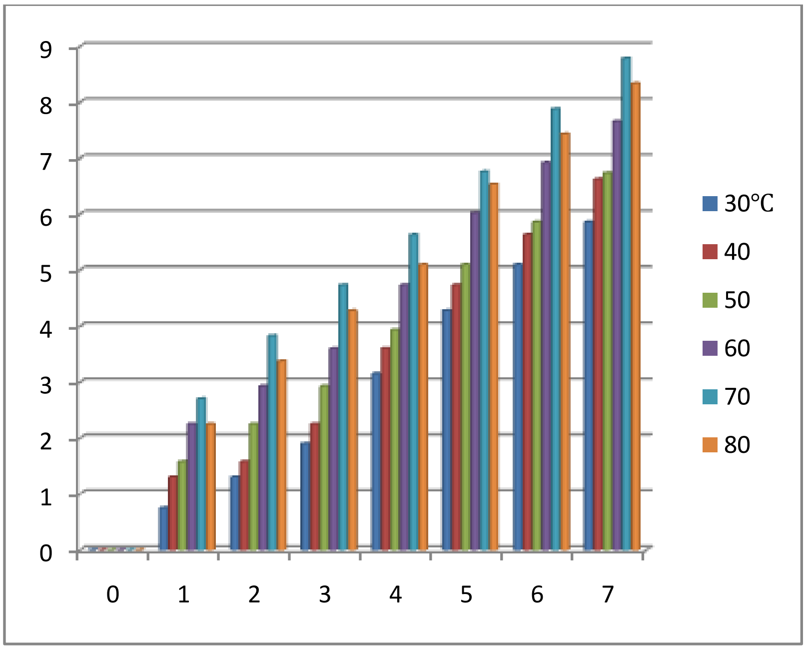
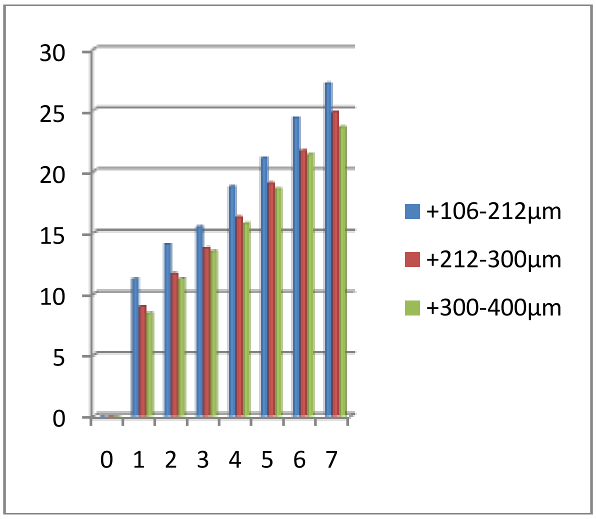


(c)

Figure 2: Conc.- time data for iron leached values by Penecillumnotatium with temperature and different particle size fractions (a) represent particle size fraction of +106-212µm; (b) particle size fraction of +212-300µm; (c) represent size fraction of +300-400µm



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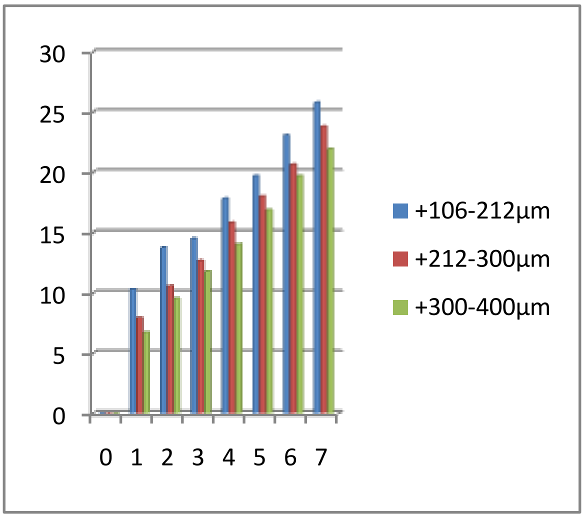

(b)

Figure 3: % of Iron Conc. Leached at 70^{o}C Vs Time for all the Sand Fractions; (a) represent bioleaching by Aspergillusniger; (b)representbioleaching by penecillumnotatium

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