



Bioaccumulation of iron metal by some fungal species

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Abstract- During the last two decades, extensive attention has been paid on the management of environmental pollution caused by hazardous material such as heavy metal. Decontamination of heavy metals (Fe, Zn, Cd, Co, Ni, Ag), in the soil and water around industrial plants has been a challenge for long time. Present studies deal with the removal of iron (Fe) metal from aqueous solutions has investigated in this study using spectronova-quantum instrument. The innovative process involved the abstraction of iron metal ions onto fungal biosorbents, followed by the application of bioaccumulation. When compared with commercial ion-exchange method, precipitation, reverse osmosis, fungal derivatives generally perform well. Fungal biomass largely depends on parameters pH, temperature, incubation periods and effects of some pretreatments. Basically this research paper provides a selective overview of present scenario of biosorption studies carried out on some promising fungal biosorbents (*Aspergillus niger* and *Penicillium chrysogenum*), which could serve as an economical means.

[Tiwary Avanti. **Bioaccumulation of iron metal by some fungal species**. *Biomedicine and Nursing* 2022;8(2):19-28]. ISSN 2379-8211 (print); ISSN 2379-8203 (online). <http://www.nbmedicine.org> 4. doi:[10.7537/marsbnj080222.04](https://doi.org/10.7537/marsbnj080222.04).

Key words: *Aspergillus niger*, *Penicillium chrysogenum*, Bioaccumulation, Biosorption, Heavy metals Microbial biomass, Metal removal.

1. Introduction

Sudden boost in industrial activities has contributed quantitatively to the alarming increase in the discharge of metal pollutants into environmental sink, especially the aqueous environment. Industrialization in India gained a momentum with initiation of five year developmental plan in the early 50's. Dispersion of the metal ions in the water bodies leads to their biomagnifications through the food chain and results in increased toxicity. The pollutants of concern include lead, chromium, mercury, uranium, selenium, zinc, iron, arsenic, cadmium, gold, silver, copper and nickel. These toxic materials may be derived from mining operations, refining ores, sludge disposal, fly ash from incinerators, the processing of radioactive materials, metal plating, or the manufacture of electrical equipment, paints, alloys, batteries, pesticides or preservatives. Metals discharged into water bodies are not biodegraded but undergoes

chemical or microbial transformations, creating large impact on the environment and public health.

Iron is applied for commercial purpose and is produced in amounts of 500 million tons annually and 300 million tones are recycled. Iron and aluminum waste products containing iron, are discharged on surface water in the earlier days. Seawater contains approximately 0.5-3 ppb of iron. Rivers contain approximately 0.5-1 ppm and groundwater contains 100 ppm of iron. Drinking water may not contain more than 200 ppb of iron. The amount of iron in drinking water about 2mg/ L does not present a hazard to health (WHO guideline for drinking-water quality, 2005). High amount of iron increasing in drinking water may cause several kind of health harm in animals, plants, human and other organism also. Iron dextrans complex repeatedly injected subcutaneously or intramuscularly was considered by IARC (International agency for Research on cancer, 1987) to be carcinogenic to animals.

In bioremoval processes, bioaccumulation plays vital role for all these purposes. Bioaccumulation is a process that uses living biomass to sequester toxic heavy metals and is particularly useful for the removal of contaminants from industrial effluents. It is a cost effective and highly specific process. This ensures reusability of biomass with short operation time. "Bioaccumulation can be defined as the ability of biological material to accumulate heavy metals from waste water through metabolically mediated or physico-chemical pathway of uptake." (Fourest and Roux, 1992).

This reaserch paper deals the bioaccumulation of iron metal by *Aspergillus niger* and *Penicillium Chrysogenum*.

2. Materials and methods

Present work deals with the culture of both the fungi *Aspegillus niger* and *Penicillim Chrysogenum* to see the bioremoval process, that is bioaccumulation for iron metal. So essential materials and apparatus, which are needed are-

2.1 Apparatus

Spectronova-quant merk is an instrument, which analyzes the concentration of any metal, which are specific for any metal analysis. A specific kit is provided with the instument. Kit having a reagent, acide thioglycolique ammonium thioglycolate with the instrument for the analysis of Fe^{++} .

2.2 Media and Reagents

Doubly distilled deionized water and anlytical reagent grade chemicals were used. $FeCl_3$ is required for the preparing the Potato Dextrose media, which is an iron containing media. 2 ppm iron containing media is prepared (Dissolve 2 mg $FeCl_3$ in 1000 ml of PDB). For maintaining pH and providing pretreatments hydrochloric acid (HCl) 1M, sodium hydroxide (NaOH) solution 1M, sodium hydroxide solution (NaOH) 1 N, hydrochloric acid (HCl) 1 N preapred. For homogenization of spores in in PDB 0.1% tween-80 also prepared.

2.3 Preparation of biosorbents

Aspergillus niger and *Penicillium chrysogenum* were grown in Potato dextrose agar media and preserved in 4°C. temperature.

2.4 Methodology

1. This experiment involved the use of potato dextrose broth (PDB) incorporated with Fe^{++} . The flasks containing media (100 ml) were subjected to initial analysis for control reading by spectronova-quant instrument.
2. The pH of flasks was maintained 6 with 1N HCl/NaOH and inoculated with the fungal culture. The culture has been obtained in PDB for 3-4 days and it has been shaken properly with 0.1% tween -80 detergents, which homogenizes the fungal spores, this suspension has been inoculated in each flask in 5 ml quantity.
3. Now flasks are used at different level for various parameters that is incubation period, pH, temperature, effects of pretreatments and dry weight of biomass and all findings are studied.

2.5 Analysis by spectronova-quant

5 ml filtrate is taken in a sterilized test tube. Kit component provided that is Acide thioglycolique ammonium thioglycolate with the instrument for the analysis of Fe^{++} , added 3 drops in the tube containing filtrate. Incubate the tube for 3 minutes. This filtrate is taken in a cuvette and analyzed in the Spectronova-quant. It shows the concentration of Fe^{++} , present in filtrate in mg/L. The readings are noted and comparative analysis between the control flasks and between the various parameters are done.

4. Results and Discussion

The bioaccumulation of iron metal using spectronova quant merk instrument by *Aspergillus niger* and *Penicillium chrysogenum* has performed with various affecting factors. The process can be understood by these results and interpretation.

3.1 Effects of incubation period (in days) on absorption-

In case of the factor incubation period, the maximum and minimum absorption was evaluated for *Aspergillus niger* at 5th day and 9th & 11th day respectively, which are noted as 3.51% and 24.7%. For

Penicillium chrysogenum the maximum and minimum absorption was observed at 5th day and 11th day respectively, which was evaluated as 4.24% and 24.58% this is because the rate of biosorption increases as the incubation increases (Fig 1).

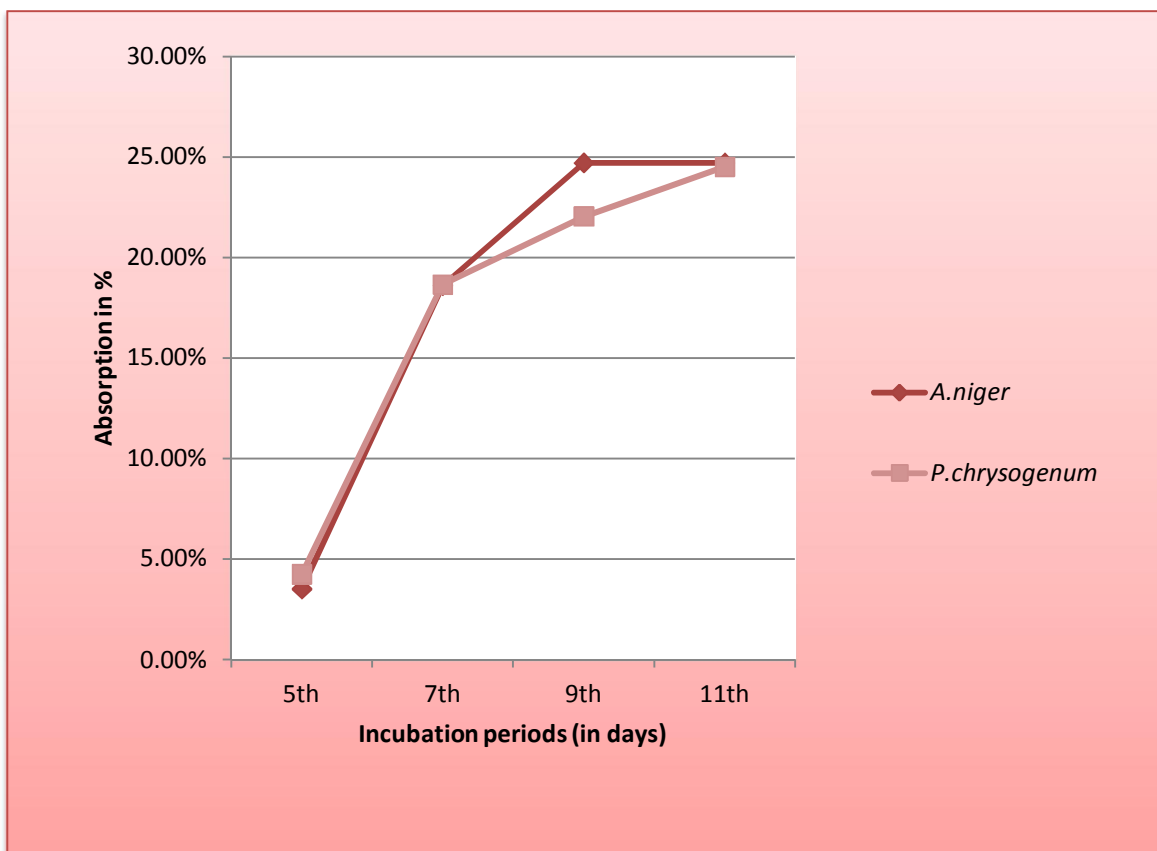


Fig: 1 Effects of incubation period on the absorption of iron by *A. niger* and *P. chrysogenum*.

3.2 Dry weight of biomass with incubation periods (in days)

Dry biomass weight changes with the incubation periods, is an important attempt to study about the mechanism. For *Aspergillus niger*, it has found that the heaviest dry weight was found at 11th day which has been noted as 0.3817g. While the lightest dry weight was found at 5th day, that was 0.2377 g. The condition was same for the *Penicillium chrysogenum* that, 11th day's dry weight was heaviest with 1.8792g. And 5th day dry weight was noted as

lightest as 1.3977g (Fig-2). These findings are noted with the incubation periods which also show the increasing manner of absorption of iron. Biosorption is a biomass surface dependent mechanism. The polymeric structure of biomass surface exhibits a negative charge due to the ionization of organic and inorganic groups and higher the biomass electronegativity, the greater the attraction and absorption of heavy metal cations. (Bux and Kasan, 1994).

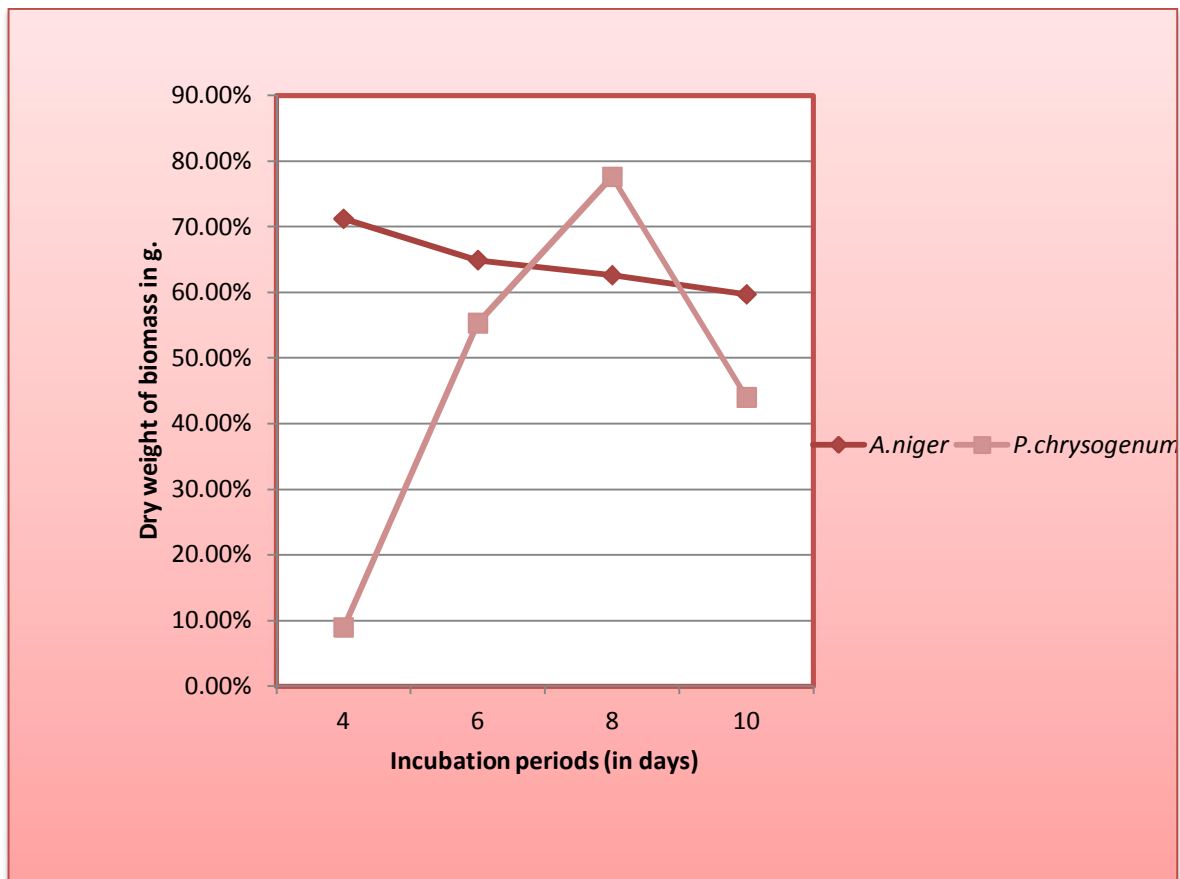


Fig: 2 Effects of incubation period on the dry weight of *A. niger* and *P. chrysogenum* during absorption of iron.

3.4 Effect of temperature on absorption-

Different status of temperature is also could be an important attempt to study about the biosorption variations. For *Aspergillus niger*, the maximum and minimum absorption was observed at 28° C and 35° C, which was gradually noted as 67.38% and 5.68%. Similarly in case of *Penicillium chrysogenum*, the maximum and minimum absorption was observed at

28° C and 35° C , which was noted as 55.3% and 2.24% respectively. According to the finding the optimum temperature for the absorption of iron in both the fungal species could be said 28° C (Fig-3). In this case temperature seems not to influence the biosorption performances in the range of 20-35 degree C. (Aksu *et al.*, 1992).

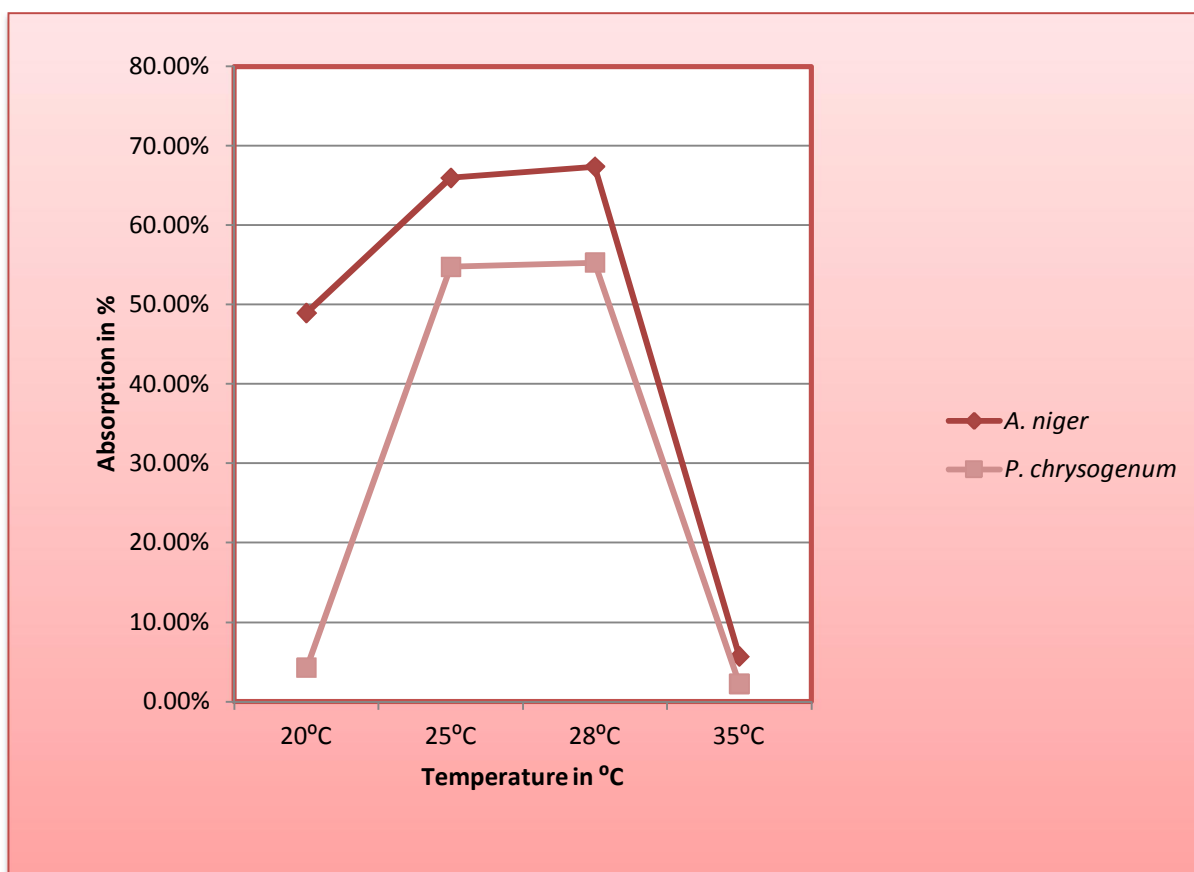


Fig 3: Effects of different temperature (in °C.) on absorption of iron by *A.niger* and *P.*

chrysogenum.

3.5 Effect of pH on absorption-

pH seems to be the most important parameter in the biosorptive process. It affects the solution chemistry of the metals, the activity of the functional groups in the biomass and the competition of metallic ions. In this work the absorption variation of iron for *Aspergillus niger* has been seen as the highest absorption was noted in acidic range at 4.0, which is evaluated as 71.2% absorption and lowest absorption was observed as pH increases and at 10.0, that is observed as 59.7%. In case of this fungus, it can be interpreted as biosorption on various fungal strains was pH sensitive. *A. oryzae*, *A. niger*, *F. solani* and *candida utilis* were found to perform better in acidic range. The

change in the sorption capacity with pH can be explained on the basis of proton-competitive adsorption reactions. (Barrous *et al.* 1998). Surprisingly in fungus *Penicillium chrysogenum* the highest absorption has been found in basic range between 6-8 pH, which was found as 77.6% while in contrast the lowest absorption has been found as 8.96% at pH 4.0. (Tan and Chang, 2003). In case of *Penicillium chrysogenum* it can be interpreted that at high solution pH values, the metal speciations in solution may become an important factor. The increase in heavy metal uptake has been attributed to reduced solubility and metal precipitation. (Barrous *et al.* 1998). (Fig 4)

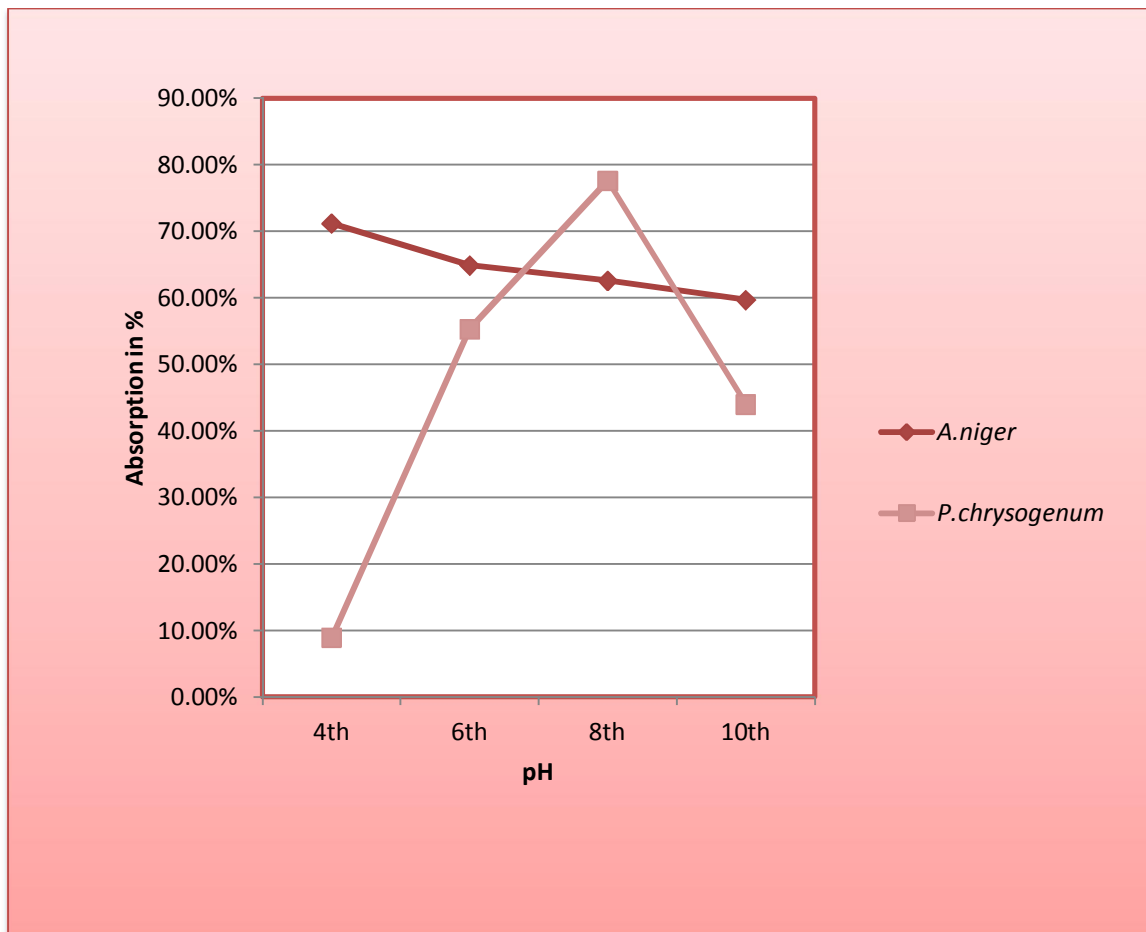


Fig 4: Effects of different pH on absorption of iron by *A. niger* and *P. chrysogenum*.

3.6 Effect of pretreatments:

Pretreatments can alter the binding affinity to metal on the biomass surface. Basically it manipulates the chemistry of biomass surface, which leads to changes in affinity towards metal. In case of *Aspergillus niger*, acid pretreatment by 1 M HCl reduces the absorption of iron as compare to biomass absorption without any treatments. In contrast, pretreatment with alkali as 1M NaOH increases the absorption of iron. In without treatments biomass the absorption has been noted as 18.6%, while in alkali and acid pretreatments the absorption has been noted as 13.27% and 34.53% respectively. In case of *Penicillium chrysogenum*, without treatment to the biomass the absorption of iron was evaluated as, 15.04%. After the alkali treatment by 1 M NaOH ,the absorption was

25.66% and after acid treatment by 1 M HCl, the absorption was noted as 37.16%. (Fig-5).

Actually the treatment with alkali usually by 1 M NaOH, deacetylates chitin present in cell wall to form chitosan-glucan complex with higher affinity for metal ions. (Muzzarelli *et al*, 1980). In contrast in the case of *A. niger*, the binding of H⁺ ions to the biomass after acid treatment may be responsible for the reduction in adsorption of heavy metals. (Kapoor and Viraraghavan ,1998). The polymeric structure of biomass surface exhibits a negative charge due to the ionization of organic and inorganic groups. Higher the biomass electronegativity, the greater the attraction and adsorption of heavy metal cations. Thus the remaining H⁺ ions on the acidic pretreated biomass may change the biomass electronegativity, resulting in a reduction in bioadsorption capacity (Bux and Kasan, 1994).

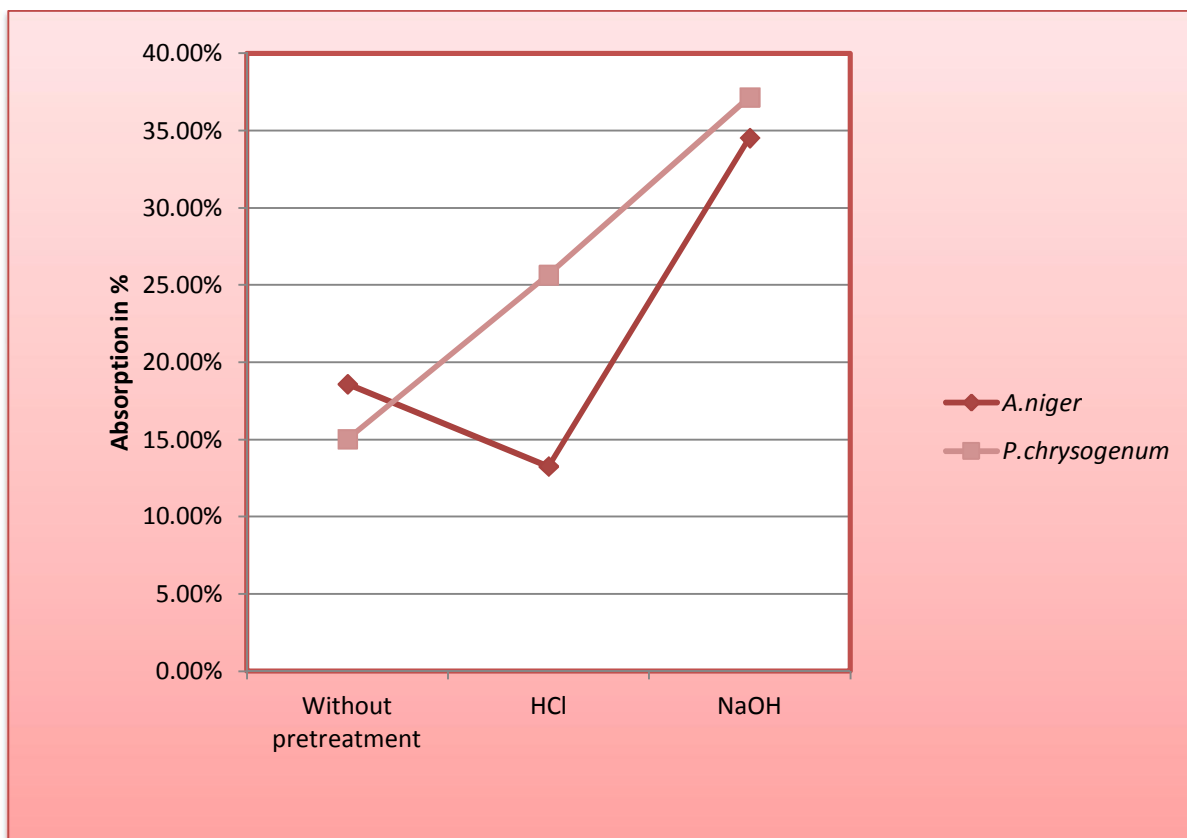


Fig 5: Effects of different pretreatments in absorption of iron by *A.niger* and *P.chrysogenum*.

5. Conclusion:

Biaccumulation is being demonstrated as a useful alternative to conventional systems for the removal of toxic metals from industrial effluents. This study was conducted to investigate the removal of iron by fungi *Aspergillus niger* and *Penicillium chrysogenum* by bioaccumulation process. The effects of various experimental parameters including incubation period, dry weight of biomass, temperature, pH and pretreatments were examined.

Collectively, using all the parameter, the optimum conditions of absorption of iron by *Aspergillus niger* and *Penicillium chrysogenum* has been investigated, which can be observed above in results. Lastly it is believed that this process offers itself as a serious alternative for a potentially extremely cost effective process for detoxification of industrial metal bearing effluents.

The indication obtained so far by a growing number of researcher present good evidence about certain types of microbial biomass possessing an exciting propensity to effectively sequester heavy metals. Bioaccumulation offers itself as a serious alternative for potentially extremely cost effective process for detoxification of industrial metal-bearing effluents.

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