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Gold Bio-recovery from Electronic Waste Using *Aspergillus niger*: A Case Study

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Abstract

Introduction: Recycling electronic waste with microorganisms is more sustainable than conventional methodologies. The usage of fungi like Aspergillus niger is comparatively sustainable for metal recovery.

Case Presentation: Aspergillus niger can be utilized for the bio-recovery of gold from electronic wastes. The study compared the efficacies of Aspergillus niger strain MXPE6 against a collection of its strains. Bioleaching was performed on printed circuit boards (PCBs) of mobile phones. The fungal consortium of strains containing MXPE6 and MX7 was found to be comparatively advantageous.

Analysis and Discussion: Aspergillus niger is highly effective at bioleaching gold. Various parameters related to fungal growth were different in the presence of PCBs. However, PCB does not interfere with fungal growth.

Conclusion: Using filamentous fungi has the potential to be efficient for metal recycling.

Keywords: Aspergillus Niger; Gold Bioleaching; Metal Bio-Recovery; Biotechnology

Introduction

Sustainably recycling electronic waste (e-waste) is extremely important. Electronic waste contains hazardous compounds that adversely impact the environment [1]. Furthermore, it contains a large number of precious metals with depleting resources such as gold, silver, copper, and platinum [2]. Electronic waste is generated in increasingly higher amounts, according to global electronic waste monitors [3]. According to the United Nations Institute for Training and Research (UNITAR), in the year 2019, 21% more electronic waste was generated globally within five years. It is estimated that the amount of electronic waste will be doubled by 2030 [4]. Hence it is important to recycle electronic waste in a method with the least negative impact on the environment [5]. Electronic wastes are recycled using physical and chemical methods [6]. Physical methods are commonly used for recycling e-waste. It involves dismantling and reducing the size of all components in the waste, after which metals can be separated. Despite this, metal recovery usually does not involve physical methods [7]. Recovery of metals from the waste is achieved using pyrometallurgical and hydrometallurgical processes [8]. Pyrometallurgical processes involve high temperatures while hydrometallurgical processes involve treatment with acids or alkalis. The hydrometallurgical method is more efficient than the pyrometallurgical method. However, both processes adversely affect the environment. Thus, biotechnological methods are considered for metal recovery from e-waste. Biotechnological methods of recovering electronic waste involve the usage of various microorganisms. This is known as biohydrometallurgy. The microorganisms are used to perform certain reactions that remove the metal component from the waste. The common methods by which microbes recover metals are bioleaching and biosorption. Bioleaching refers to the change in a metal compound into a soluble compound. Biosorption refers to the concentration and binding of ions on biological surfaces. In some cases, the ions can be concentrated inside the biomass, which is known as bioaccumulation.Bacteria are commonly used for the bioremediation of e-waste. Bacteria recover the metals generally by bioleaching. The bacteria produce certain compounds during metabolism that enable the bioleaching of the metal. These compounds are called lixiviants. Iron-oxidizing bacteria, sulphur-oxidizing bacteria, cyanogenic bacteria, and organic acid-producing bacteria are typically utilized for metal recovery. However, several fungi can be used for this purpose as well.The fungal species that are normally used for this purpose *are* Penicillium simplicissimum and Aspergillus niger. These fungi produce organic acids that attack mineral surfaces as well as act as lixiviants. Recovery of metals is controlled by the solubilization of metal from the waste by these lixiviants. Bioleaching by lixiviants occurs through numerous mechanisms such as acidolysis, complexolysis, redoxolysis, and alkaloysis. Several fungi can also immobilize the gold through biosorption, bioaccumulation, and biomineralization. Fungi are preferable for metal recovery for a few reasons. Firstly, the usage of fungi is relatively more sustainable than certain bacteria due to lesser environmental risk. This is due to the production of organic acids by the fungi. This paper is a case study on the usage of *Aspergillus niger* strains for gold recovery from printed circuit boards. There have been similar studies on gold recovery by fungi. However, in this study, efficient bio-recovery of gold with lower production costs is depicted.

Case study on gold remediation

A study was conducted in the year 2019 in Mexico to check the performance of Aspergillus niger strains in the bioremediation of gold. The study was performed by Rosalba Argumedo-Delira, Mario J. Gómez-Martínez, and Brenda Joan Soto. The research involved analysis of bioleaching of gold by Aspergillus niger only. Strain MXPE6 was compared against a fungal consortium composed of MXPE6 and MX7 strains. The bioremediation was performed on PCBs (printed

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circuit boards) taken from mobile phones. The distinguishing features of this study were that it employed glucose as a carbon source and the culture medium was not agitated.

Choice of Aspergillus niger Strains

The *Aspergillus niger* strains involved in the study were strains MX7 and MXPE6. Both strains were isolated from a landfill in Tronconal, Xalapa, Veracruz, Mexico. MX7 was isolated from the soil around the landfill. On the other hand, isolation of MXPE6 was accomplished using an electronic board from the same place. The strains taken for research thrived on metals including gold. Thus they have been obtained in metal-contaminated or metal-rich environments. The selection of both strains was based on a study conducted by Madrigal-Arias et. al. This study exhibited the differences between MX7 and MXPE6 strains in terms of gold bioleaching. In the study, strain MX7 was said to possess less efficacy in gold bioleaching compared to other gold-tolerant strains. Furthermore, strain MXPE6 showed comparatively higher tolerance to gold. Thus, the MX7 strain was taken as a part of a mixture of strains. When the bioleaching ability of MXPE6 only against two strains of *Aspergillus niger* including MX7 were evaluated, it was found that two strains were more effective. Gold bioleaching amount by both MXPE6 and MX7 was 28 times higher than the amount of gold bioleached by MXPE6 inoculation. Besides, differences between both strains were apparent in the given study as well. MX7 grows within five days in a Petri dish containing potato dextrose agar grown at 25 ^{0}C. Whereas, MXPE6 grows slower than MX7, taking a week to grow in the same environmental conditions with yellow pigmentation.

Usage of the Electronic Waste

The PCBs taken from the mobile phones were used to prepare the culture medium. The PCBs were recovered from the mobile phones by using physical methods and chemical treatment. The physical methods involved dismantling the mobile phones and preparing a powder out of the PCBs. After sterilization and drying, four sample solutions were prepared. 50 mL of sample solution was prepared to check gold content in the PCBs using optical emission spectrometry. The powder was sterilized using ethanol for four hours. Furthermore, for sample digestion, aqua regia solution was used. The sterile PCB powder was used as a component of the culture medium. The composition of the culture medium is 1.6 L mineral media at pH 4.4 and eleven grams of powder. The mineral media was made up of ammonium sulphate, sodium dihydrogen phosphate, calcium chloride, magnesium chloride, and glucose.

Growth of Strains

The strains were grown in plastic bioreactors with a 5 mL capacity. After the growth of strains on Petri plates, Thirty-eight disks with fungal mycelia were put inside the bioreactor for both MXPE6 and fungal consortium. Nineteen disks containing mycelia of only one fungal strain were used as well; each strain possessed its bioreactor. Later, two control bioreactors were constructed as well. The first control bioreactor contained PCB-free media while the second control bioreactor had a control reactor without fungal inoculum. The bioreactors were incubated for 32 days for strain cultivation. It was cultivated and monitored at room temperature. Stoichiometric coefficients were calculated to analyze strain growth. The stoichiometric coefficients evaluated were fungal growth rate, coefficient of biomass yield, and coefficient of gold bioleaching per glucose consumed. Three samples were collected. Sample collection was performed every 96 hours for monitoring the following variables: total sugar present, pH, fungal protein content, total dry biomass, and the amount of gold leached.

Analysis of Fungal Biomass

The fungal growth obtained from the bioreactor was analyzed. The control without PCB, called the biotic control, was compared with the sample bioreactor for comparison of concentrations of glucose and proteins as well as total biomass yield. Firstly, analysis was performed to evaluate the effect of PCB on glucose utilization by biomass. It was found that the presence of PCB did not affect the utilization of glucose since it was similar in both cases. It was also performed to examine the effect of PCB on protein concentrations within the biomass. There was an increase in protein content only within the MXPE6 strain. Secondly, analysis was performed to check biomass yield. Biomass yield was highest for fungal consortium without PCB. Furthermore, the result of the analysis indicated that the presence of PCB resulted in a marginally higher fungal growth rate. It also showed that the total dry biomass was significantly higher in the media with PCB.

Bioleaching of Gold by Strains

Bioleaching of gold by the samples was studied by analysis of certain parameters. They were gold bioleaching coefficients, changes in pH, and gold accumulation by biomass.

Gold bioleaching by the MXPE6 and the fungal consortium were significantly different. The fungal strain bioleached more gold than the MXPE6 strain. However, the calculation of the gold bioleaching coefficient showed that MXPE6 bioleached three times the amount of gold than the fungal consortium. Furthermore, the fungal consortium sample possessed lower pH than the single strain, indicating a higher number of organic acids released. This was an indicator that the fungal consortium bioleached more gold. Moreover, there was 10% less gold accumulation by the MXPE6 strain

Analysis and discussion

The Action of *Aspergillus niger* on Printed Circuit Boards (PCBs)

Understanding the reason for the usage of Aspergillus niger for gold recovery from printed circuit boards (PCBs) involves analyzing its effects. Filamentous fungi like Aspergillus niger are used for metal recovery for various reasons. Filamentous fungi produce organic acids that help in metal removal. The filamentous nature of the fungi enables them to survive in heterogeneous and solid environments. They are extremely tolerant to various environmental stresses. Some filamentous fungi have primary resistance to metals because of their cell wall. Moreover, filamentous fungi can bioleach metal faster since they can reach the exponential phase of growth faster and produce the primary metabolites for bioleaching. All these factors enable better bioleaching of precious metals like gold.

Aspergillus niger can be used for the recovery of many metals at the same time. This is particularly useful in performing bio-recovery of a large amount of metal. Furthermore, PCBs consist of numerous metals, including gold, copper, silver, nickel, and platinum. This makes the usage of Aspergillus niger on PCBs particularly advantageous. However, in the study, it is only used for recovering gold. The reason behind this is that *Aspergillus niger* was found to have the highest capability in gold bio-recovery. A study was conducted on the bioleaching of gold, silver, and copper from PCBs of mobile phones by different microorganisms. It showed that Aspergillus niger performs the highest bio-recovery of gold. Moreover, MXPE6 and fungal consortium with it showed high gold leaching percentages.

Effect of Electronic Waste on *Aspergillus niger*

In the study, there were different values for fungal growth in biotic control and bioreactor with PCB present. The parameters that were affected were growth rate per glucose consumed, biomass yield coefficient, and amount of dry biomass obtained. However, the correlation between the presence of PCB in the culture medium and fungal growth was not found. In the case of growth rate, there was a change in growth rate only when MXPE6 was inoculated with PCB. There were no changes in the fungal consortium growth rate, only an increase in the growth rate of MXPE6. In the case of biomass yield, it increased in presence of PCB for single inoculum and decreased in the case of fungal consortium. Total dry biomass was significantly higher in the presence of PCB. Upon correlation of these parameters with the presence of PCB, it was found that all parameters, while being correlated to each other, were not correlated to it. The presence or absence of electronic waste material (PCB) did not affect the correlational values. Hence, e-waste does not affect the growth or bioleaching capabilities of Aspergillus niger.

Conclusion

The usage of filamentous fungi is a sustainable method of metal bio-recovery. Metal bio-recovery is important in recovering precious, rare metals in a sustainable method. They possess less environmental risk and can perform metal bio-recovery efficiently. They possess the capability of removing multiple metals from the same solid. They are highly adaptable, and their growth is not affected by the presence of electronic waste. They also perform bioleaching of metals faster than certain bacteria. Aspergillus Niger, in particular, is highly efficient in the remediation of gold and copper. Fungi like *Aspergillus niger* have been studied for large-scale metal recovery processes. However, only experiments for metal recovery have been conducted. This is because it is time-consuming, less efficient, and more expensive compared to conventional methods of metal recovery from e-waste. Currently, the usage of bio-metallurgy is not as developed as physical or chemical methods. Hence more research is required for developing large-scale recovery of metal from e-waste.

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