

# BIOSORPTION OF COPPER (II) BY IMMOBILIZED *ASPERGILLUS NIGER*

**J. JAYACHITRA**, Assistant Professor  
Department of Agricultural Microbiology  
Faculty of Agriculture, Annamalai University, Annamalai Nagar

**E. BABU**, Assistant Professor  
Department of Chemical Engineering,  
Faculty of Engineering & Technology  
Annamalai University, Annamalai Nagar – 608 002

## Abstract

A biological material for the removal of heavy metal ions from waste water is important due to its extreme toxicity towards aquatic life and humans. Micro organisms are being increasingly studied for the removal of heavy metal ions from aqueous solution. In this work, *Aspergillus Niger* powder was taken as a biosorbent for the sorption of Copper(II). The various parameters like initial metal ion concentration, initial pH, temperature and biosorbent dosage were studied in a batch reactor. Equilibrium was reached after 24 h of contact time. The optimum values of initial copper concentration, initial pH, temperature and biomass loading are found to be 50mg/l, 4.30°C and 5g/l. Under this optimised condition, a maximum percentage removal of 91% and specific uptake of 11.2mg/g was obtained for Cu (II) sorption.

**Key words:** Biosorption, *aspergillus Niger*, initial concentration, percentage removal, uptake of metal ion, etc.

## 1. Introduction

Environmental contamination with heavy metals became a worldwide problem due to the risks they can generate for ecosystems and human health. Many heavy metals such as lead, mercury, nickel, arsenic, chromium, zinc, copper, cadmium, cobalt, antimony, etc. in their elemental forms or in various chemical combinations are considered as toxic. They are non-degradable toxic pollutants have a great tendency to bio accumulate; some are carcinogenic or have mutagenic effects for human and can seriously affect the living organisms. In this context, different studies were developed on the removal of toxic heavy metal from contaminated media. The most widely used methods are: the conventional physico-chemical processes, ion exchange, adsorption on activated carbon, etc. Some of these methods are relatively expensive unfriendly for the environment and their effectiveness may depend on the concentration of pollutants in the contaminated environment. Moreover, these methods are associated with high energy consumption or an incomplete removal of the pollutant. Therefore numerous studies are conducted for the development of cheaper and more effective technologies. In this context, a biological method like biosorption is considered as good, efficient and cost effective. Since the use of biomass a very cost effective source of biosorbents with high uptake capacity, these methods are considered eco friendly and economic options with high efficiency for removing heavy metals from waste waters. Among the heavy metals, copper is one of the most important heavy metal which is extensively used in most of the industries. This present study includes removal of copper from aqueous solution of Cu (II) by fungal strain *Aspergillus Niger* MTCC 132.

## 2. Materials and methods

### 2.1. Preparation of sorbate solution

A 1000 mg/l stock solution of copper was prepared by dissolving 3.93 g of copper sulphate in double distilled water. The required concentrations of copper ions were prepared from the stock solution by dilution method.

### 2.2 Preparation of the biosorbent

*Aspergillus Niger* culture from agar slants is transferred to 50 ml of liquid medium containing potato extract, dextrose and yeast extract. The culture is grown initially in filamentous form with white fluffy mycelia and finally it is matured to dark colonies with black spores. At the deceleration phase of the growth, the contents of the vessels are harvested by filtering through nylon mesh. The recovered biomass is washed extensively with tap water. After washing, the biomass is dried at 60°C for 12-15h and powdered using mortar and pestle. The powdered biomass is sieved through a sieve with openings of 75 µm and the undersize particles are used in immobilization. The oversize particles are used as suspended dried cells for subsequent use.

### 2.3 Preparation of immobilized biomass beads

Immobilized biomass beads are prepared using 8% (w/v) sodium alginate. A known amount of biomass (*Aspergillus Niger*) is mixed with sodium alginate and the mixture is constantly stirred under warm condition until the alginate gets dissolved. The suspension is dripped into 2% (w/v) calcium chloride solution through a syringe. The beads are stored in calcium chloride solution for about 30 minutes before being rinsed in double distilled water.

### 2.4. Batch biosorption studies

Batch experiments were carried out in Erlenmeyer flasks by adding known amount of immobilized biomass beads in 100 ml aqueous copper sulphate solution. The flasks were agitated on a shaker with a constant shaking rate at 150 rpm for 240 min until equilibrium sorption was obtained. Samples were taken from the solution at regular time intervals for the residual metal ion concentration in the solution. The residual concentration of copper ions in the solutions was determined spectrophotometrically at 457 nm using Neocuproine as the complexing agent. The effect of initial copper ion concentration on percentage removal of copper was studied by conducting experiments with different initial copper ion concentrations namely 50 mg/l, 100 mg/l, 150 mg/l, 200 mg/l and 250 mg/l under identical conditions of temperature, pH and biomass loading and the experiment was carried out as described above. The effect of initial pH on percentage removal of copper was studied by conducting experiments with different initial pH namely 2,3,4,5 and 6 under identical conditions of initial Cu (II) ion concentration, temperature and biomass loading and the

experiment was carried out as described above. The effect of temperature on percentage removal of copper was studied by conducting experiments with different temperature namely 25°C, 30°C, 35°C and 40°C under identical conditions of initial Cu (II) ion concentration, initial pH and biomass loading and the experiment was carried out as described above. The effect of biomass loading on percentage removal of copper was studied by conducting experiments with different biomass load namely 1g/l, 2g/l, 3g/l, 4g/l and 5g/l under identical conditions of initial Cu (II) ion concentration, initial pH and temperature and the experiment was carried out as described above.

### 3. Results and discussions

The biosorption of metals using immobilized biosorbent in a batch process depends on both contact time between the adsorbate and adsorbent particles and initial metal ion concentration. The effect of initial metal ion concentration on contact time, percentage removal and specific uptake of copper was given in Figure 1 and Figure 2 respectively. Figure 1 shows that equilibrium is attained in 24 h, also the sorption of copper on immobilized biosorbent increases with increasing contact time. The copper removal efficiency was affected by the initial metal ion concentration, with decreasing removal percentages as concentration increases from 50 mg/l to 250 mg/l. As the initial copper concentration increases from 50 mg/l to 250 mg/l, the percentage removal of copper decreases from 91% to 81% and the specific uptake of copper increases from 11.2mg/g to 50.5mg/g respectively. At lower initial metal ion concentrations, sufficient adsorption sites are available for adsorption of metal ions. However, at higher concentrations, the number of metal ions relatively higher compared to availability of adsorption sites. The percentage of metal sorption varies with pH of the medium which is given in Figure 3. The percentage removal of copper increases from 76% to 92% as the pH increased from 2.0 to 4.0 and thereafter increasing pH up to 6, the percentage removal of copper decreased to 86%. Biosorption of copper was low at alkaline condition. The maximum percentage removal is found to be 92% at pH 4.0 and selected as the optimum pH. The percentage removal of copper by immobilized biosorbent was very less at low pH. The result seems to suggest that the biosorption of copper by immobilized *Aspergillus Niger* is mainly due to ionic attraction. This can be explained based on at low pH, highly mobile H<sup>+</sup> ions are adsorbed at the active sites, preventing copper ions from getting sorbed. At pH values higher than 5.0, percentage removal of copper decreases because of the increase in concentration of OH<sup>-</sup> ions in the biosorption medium causes the precipitation of copper. Hence, biosorption studies should not be carried out at higher pH levels which cause precipitation of metal ions. Optimum metal biosorption at pH 4-6 has also been reported for several other biomass types, and is likely due to deprotonation of metal binding anionic sites, such as carboxylic groups at this pH range.

The effect of temperature on percentage removal of copper was studied in Erlenmeyer flasks with 100 ml of aqueous copper solution at different controlled temperatures namely 25°C, 30°C, 35°C and 40°C. The effect of temperature on percentage removal of copper by immobilized biosorbent was

given in Figure 4. A maximum copper removal of 67% is obtained at 30°C because the number of binding sites is more at this temperature. The percentage removal of copper by immobilized biosorbent is higher at room temperature and it decreases with further increase in temperature due to the destruction of the cell walls expected, and a reduction in copper removal is observed. The effect of biomass loading on percentage removal of copper was studied by conducting the experiments in Erlenmeyer flasks with 100 ml of aqueous copper solution with different biomass loading namely 1g/l, 2g/l, 3g/l, 4g/l and 5g/l. The results of effect of biomass loading on contact time and percentage removal of copper during the biosorption process are given in Figure5. It was observed that the percentage removal of copper increased from 71 to 85% as the biomass loading increased from 1 g/l to 5 g/l. At low copper concentration, the ratio of sorptive surface to the total Cu (II) ions available is high and nearly all copper ions in solution can be bound and removed. The increase in the uptake capacities of immobilized *Aspergillus Niger* with increasing metal concentration may be due to higher probability of collision between metal ions and biosorbent particles. A maximum copper removal of 86% was observed at a biomass loading of 5 g/l.

#### 4. Conclusion

Biosorption experiments were performed as a function of initial metal ion concentration, pH, temperature, and biosorbent dosage. Biosorption was influenced by initial copper ion concentrations and it was found that as the initial copper concentration increases from 50 mg/l to 250 mg/l, the percentage removal of copper decreases from 91 % to 81 % and the specific uptake of copper increases from 11.2 mg/g to 50.5 mg/g respectively. The effect of initial pH was also influence the sorption efficiency. The optimum conditions were found to be initial concentration 50mg/l, pH 4, temperature 30°C and biomass loading of 5g/l. The obtained results showed that immobilized *Aspergillus Niger* was a good adsorbent for the removal of metal ions and had high adsorption yields for the treatment of aqueous solutions containing copper ions.

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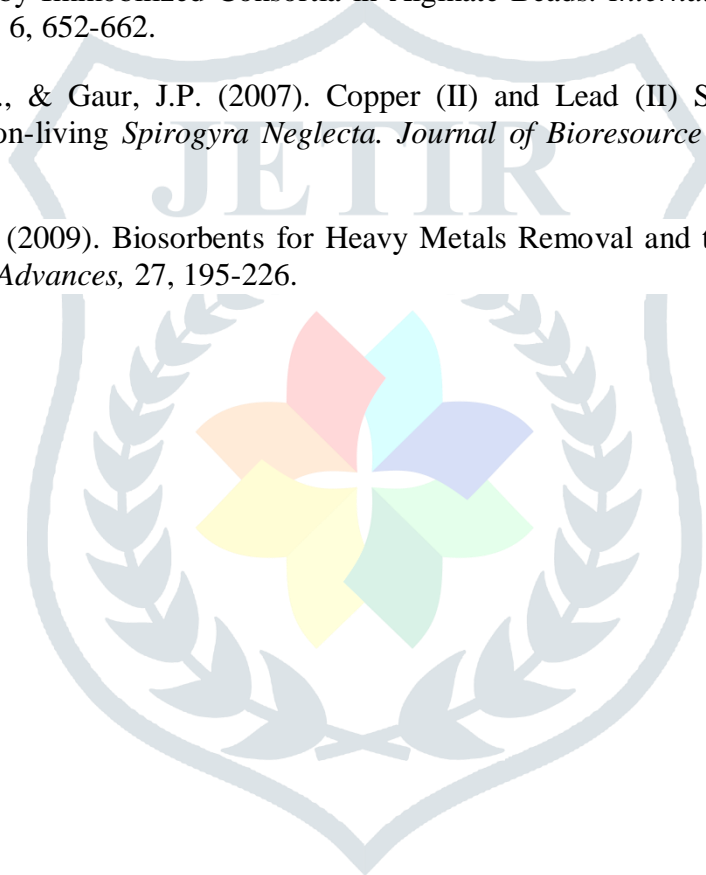
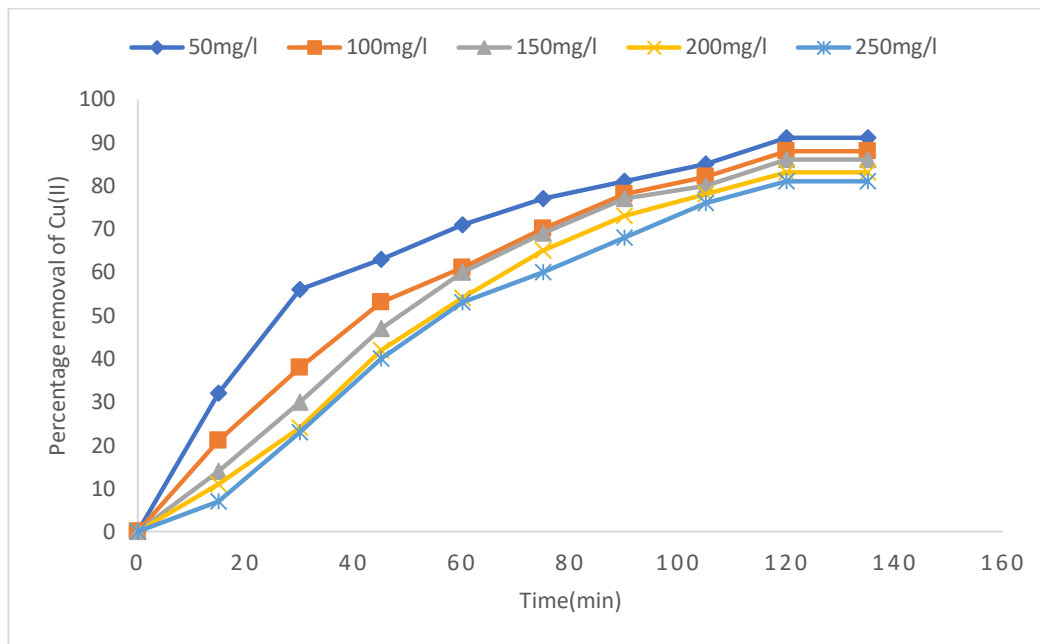


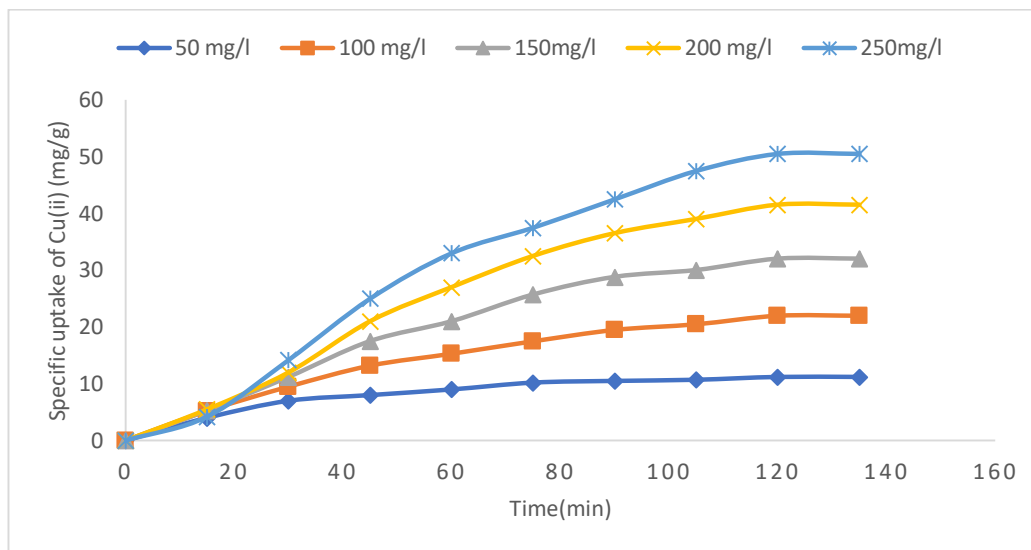
FIGURE 1

Effect of Initial Copper Concentration on Percentage Removal of Copper by *Aspergillus Niger*



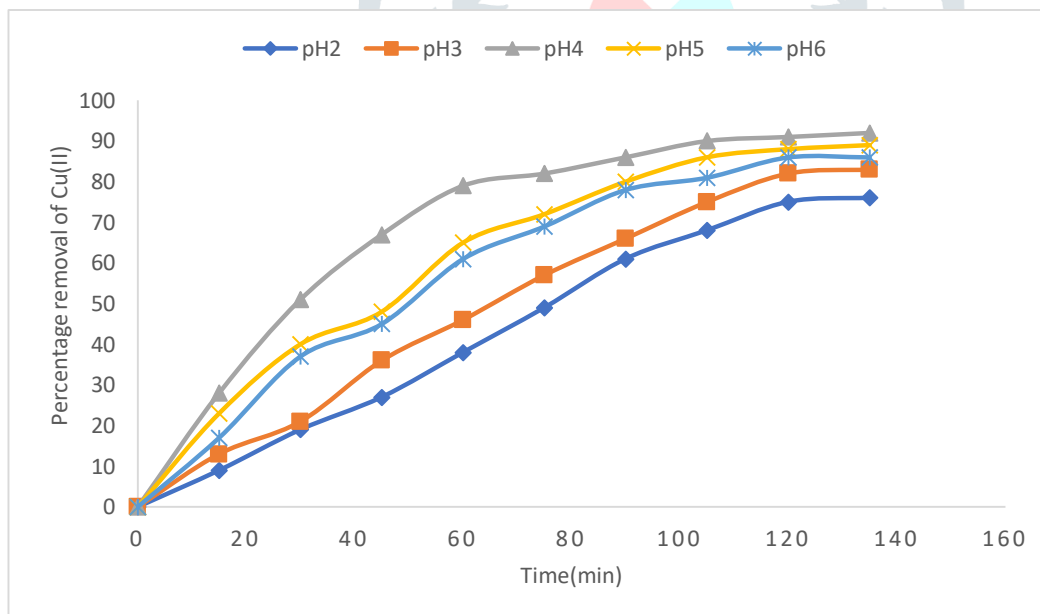
**FIGURE 2**

**Effect of Initial Copper Concentration on Specific Uptake of Copper by *Aspergillus Niger***



**FIGURE 3**

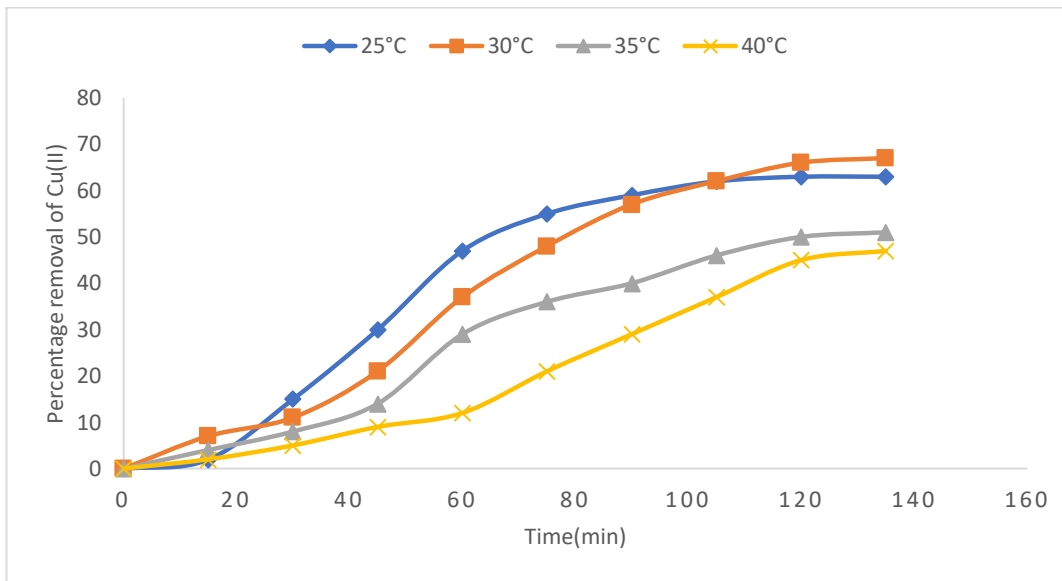
**Effect of Initial pH on Percentage Removal of Copper by *Aspergillus Niger***





**FIGURE 4**

**Effect of Temperature on Percentage Removal of Copper by *Aspergillus Niger***



**FIGURE 5**

**Effect of Biomass Loading on Percentage Removal of Copper by *Aspergillus Niger***

