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Lead(II) and Iron(II) Removal from Aqueous Solution: Biosorption by a Bacterial Biofilm Supported on Granular Activated Carbon

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ABSTRACT

A biofilm of *Arthrobacter viscosus* supported on granular activated carbon (GAC) removed between 100% and 50% of Pb(II) and between 100% and 30% of Fe(II) from solution with initial concentration between 8–85 $mg_{Pb}\ l^{-1}$ and 2.5–42 $mg_{Fe}\ l^{-1}$ and a flow residence time of 1.2 min. The maximum uptake capacities of the system biofilm/GAC ranged from 4.8 mg_{Pb}/g_{GAC} to 24.2 mg_{Pb}/g_{GAC} and 1.8 mg_{Fe}/g_{GAC} to 18.4 mg_{Fe}/g_{GAC}. The behavior of granular activated carbon with two different surface treatments (treated with HNO_3 or H_2O_2) was studied and best results were achieved with the support treated with HNO_3. The polysaccharide and polymeric net was also studied, and it was concluded that the production of polysaccharides and polymers was much higher in the GAC-HNO_3.

Keywords: activated carbon, *arthrobacter viscosus*, biofilm, biosorption, heavy metals

INTRODUCTION

Biosorption of heavy metals is one of the most promising technologies aiming at the removal of toxic metals from industrial waste streams and natural waters. It is a potential alternative to conventional processes for the removal of metals such as precipitation. The most important key factors that should be considered in the application of biosorption in the removal of toxic metals from industrial waste solution are: the use of low cost waste biomass, the cost of biomass immobilization and the possibility of a continuous re-usage of the biomass

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through sequential operation of biosorption and regeneration (Gasbarro *et al.*, 1997).

Many microorganisms are able to accumulate heavy metals from solutions. They accumulate metals by a number of different processes such as uptake by transport, biosorption to cell walls and entrapment in extracellular capsules, precipitation and oxidation–reduction reactions (Duncan and Brady, 1994). This ability to attract and subsequently sequester metals from the surrounding aqueous environment is a consequence of the presence of several chemical groups on the biomass surfaces, such as acetamido groups of chitin, amino and phosphate groups of nucleic acids, amino, amido, imino, sulphydryl and carboxyl groups of proteins (Hancock *et al.*, 1999). The use of bacteria presents some advantages like their small size, their ability to grow under controlled conditions and their resilience to a wide range of environmental situations (Urrutia, 1997).

The effect of carbon surface chemistry on the biofilm formation is an important factor that should be considered. According to Noll *et al.* (2000), four major oxidized groups are produced when activated carbon is oxidized by nitric acid: carboxyl, lactonized carboxyl, hydroxyl and carbonyl groups. In this way the granular activated carbon treated with nitric acid presented more surface functional groups with oxygen than the one treated with hydrogen peroxide.

The aim of this study was the investigation and development of an innovative process for the removal of heavy metals. *Arthrobacter viscosus* supported on granular activated carbon was chosen as biosorbent material due to the relative lack of information about its sorption properties and its evident ability to produce large amounts of exopolysaccharides that may determine the efficiency of the whole process (Scott and Palmer, 1988). The effects of the initial concentration of metal and surface treatment of carbons were tested and the polysaccharide and polymeric net were quantified.

MATERIAL AND METHODS

Materials

The bacterium *Arthrobacter viscosus* was obtained from the Spanish Type Culture Collection of the University of Valência. Aqueous lead and iron solutions were prepared by diluting $Pb(NO_3)_2$ and $FeSO_4.7H_2O$ in distilled water. All glassware used for experimental purposes was washed in 60% nitric acid and subsequently rinsed with deionised water to remove any possible interferences by other metals. Atomic absorption spectrometric standards were prepared from 1000 mg_{lead} l^{-1} and 1000 mg_{iron} l^{-1} solution. The support was characterised by

N_2 adsorption (77 K), with a ASAP Micromeriticcs 2001, in order to evaluate surface area, pore size distribution and pore volume. Surface treatments consisted in washing the granular activated carbon (GAC) for 1 h at 90°C, with a solution of 1M HNO_3 or 1M H_2O_2, in order to develop different surface groups in the carbon surface, after thermal treatment in N_2 atmosphere. Surface functional groups identification was carried out by Boehm titration, FTIR, TPD and XPS. Universidade Nova de Lisboa, Departamento de Química, made the structure and surface characterization as well as surface treatment of carbons.

Methods

Biosorption studies

All experimental work was conducted in duplicate. GAC was placed in an Erlenmeyer flask of 250 ml to which was added 150 ml of distilled water. It was sterilized at 120°C for 20 min to release the air inside the pores. Then it was placed in column. Minicolumns (internal diameter = 0,9 cm, ht = 30 cm) were used for open systems studies, partially filled with GAC (6 g) with a Langmuir aarea of 1270 $m^2.g^{-1}$ and an average pore diameter of 20 nm. The microorganism culture and the nutrient broth were pumped through the bed aiming the formation of the biofilm. Two different media, with different concentrations of peptone, were used to grow the microorganism during 3 d, aiming the optimization of the adhesion (see Tables 1 and 2). Previous studies realized by this group showed' that the use of these two different media maximized the production of polysaccharides and total polymers. The formation of the biofilm was observable by naked eye. After this period of time the bed was washed out and the metal solutions were treated passing through the column with a flow rate of 10 ml/min. Samples (5 ml) were taken, centrifuged and analyzed for metals using atomic absorption spectrophotometry, AAS. The results were expressed as removal percentage and uptake. At the end of each run the column was washed out and samples of the effluent were seeded in Petri plates with nutrient agar to assess the metabolic activity of the microorganism.

Quantification of polysaccharides and total polymers

The method used for the quantification of polysaccharides and total polymers was first described by Oliveira and Azeredo (1996). It consists of three steps: i) Solubilization of the polysaccharide and polymeric net with a solution of

TABLE 1
Composition of growth medium and cultivation conditions (during 1 day)

Component (g/l) or condition	
Malt Extract	3
Yeast Extract Powder	3
Peptone	5
Glucose	10
pH	7
T (°C)	26

TABLE 2
Composition of growth medium and cultivation conditions (during 2 days)

Component (g/l) or condition	
Malt Extract	1
Yeast Extract Powder	1
Peptone	0.167
Glucose	3.3
pH	7
T (°C)	26

glutardialdehyde, ii) Dialysis of the obtained solution, and iii) Precipitation of the dialyzed.

RESULTS AND DISCUSSION

Heavy metal removal was rapid and showed the best results during the first hour. The removal of lead and the removal of iron from the liquid phase follow a typical biosorption kinetics which includes two distinct processes. There is an initial rapid accumulation step that is described as to be independent of metabolism and temperature and is thought to involve cation binding at the surface, biosorption itself. This step is followed by a second process that appears

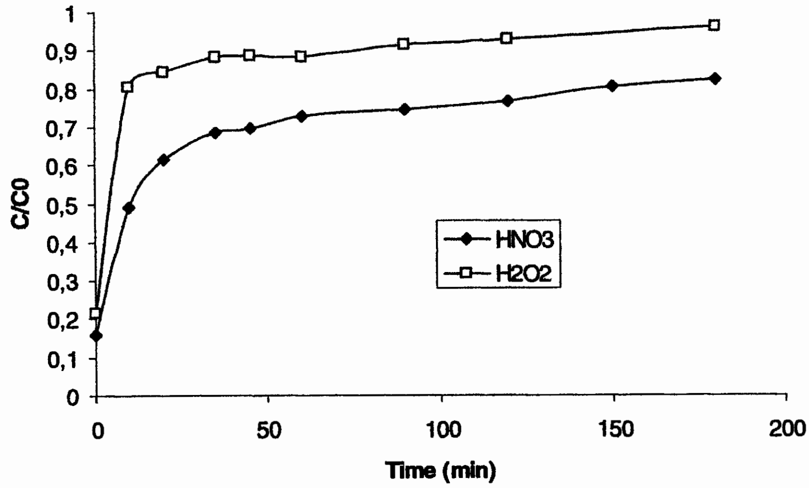


Figure 1. Breakthrough curves for lead by a biofilm of *Arthrobacter viscosus* supported on granular activated carbon, with surface treated with HNO_3 and H_2O_2. The initial concentration of lead was 30 mg l^{-1}, with a flow rate of 10 ml min^{-1} and a residence time of 1.2 min.

to be metabolism-dependent, much slower, and can accumulate larger quantities of cation than the first process (Figure 1). This second process is believed to involve cation internalization into the cell (Duncan and Brady, 1994).

The quantification of polysaccharides and total polymers show that the production of polysaccharides and polymers was much higher in the GAC-HNO_3 (Figure 2). These results presigned a relationship between biosorption efficiency and polysaccharide net. The difference between the results obtained in different experiments is a consequence of the many variables without control like quantity of microorganism present, quantity of biomass formed and the quantity of gas produced by the microorganism during fermentation, for example.

Effect of initial concentrations of metals

The removal percentage decreases with the increase of the initial concentration of metals (Figure 3 and Figure 4). The removal of Pb reaches 50% (GAC-H_2O_2) and 75% (GAC-HNO_3) with an initial concentration of 8 mg_{Pb} l^{-1}, which is reduced to 45% (GAC-HNO_3) with an initial concentration of 17 mg_{Pb} l^{-1}. In the case of Fe, the metal removal reaches 80% (GAC-HNO_3) and 73% (GAC-H_2O_2) with an initial concentration of 2.5 mg_{Fe} l^{-1}, which is reduced to 60% (GAC-HNO_3) and 40% (GAC-H_2O_2) when the initial concentration is

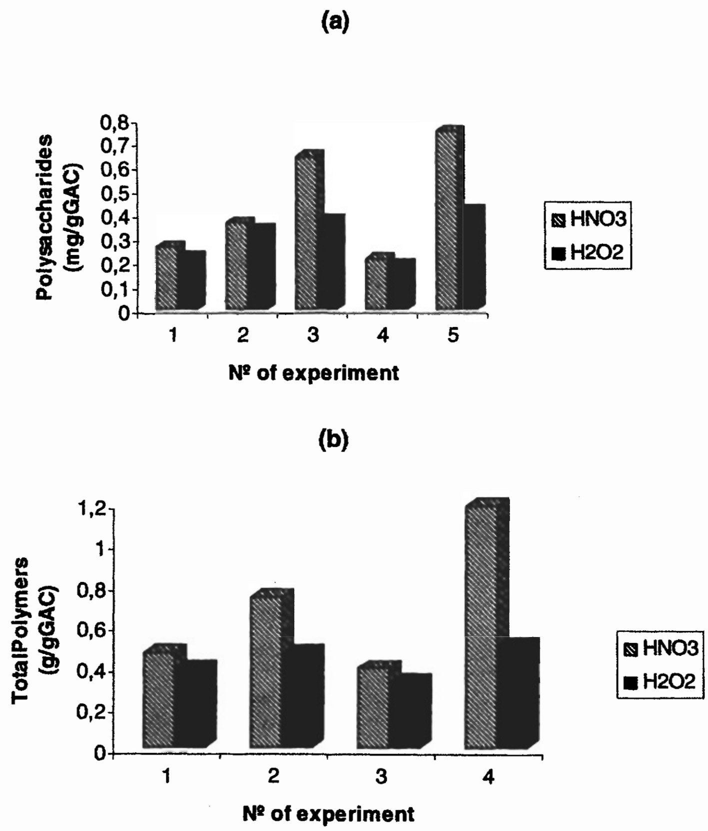


Figure 2. A comparison of the production of polysaccharides (a) and total polymers (b) by a biofilm of *Arthrobacter viscosus* supported on granular activated carbon, with surface treated with HNO_3 and H_2O_2.

increased to 5 $mg_{Fe}\ l^{-1}$. This can be explained by the saturation of the binding sites present in the cell wall and by a growing toxic effect of metals felt when the initial concentration of metals is higher. The difference between the behavior of the metals is, probably, a consequence of the elevated atomic weight of lead (that promotes the biosorption) and the reduced ionic radius of iron (that promotes the penetration in the polymeric net).

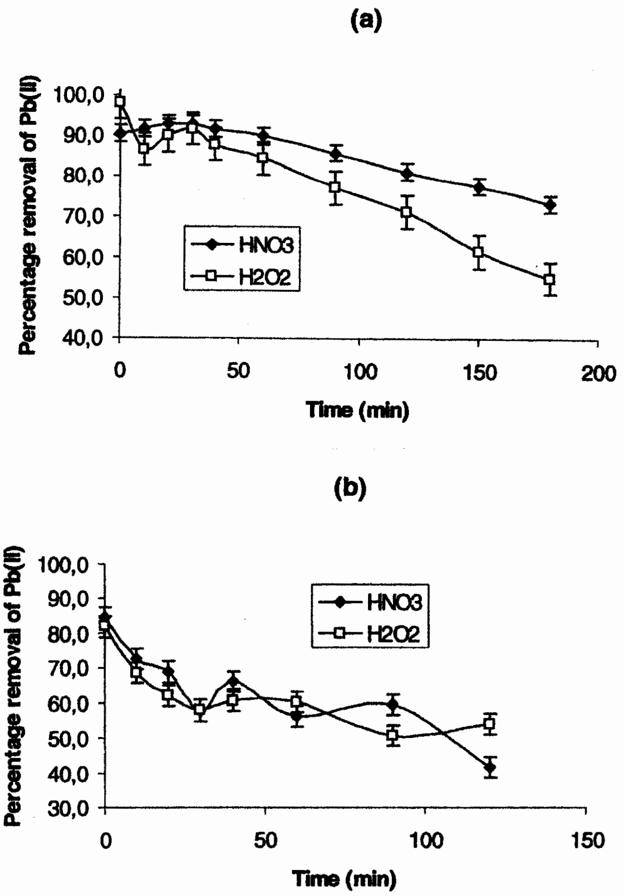


Figure 3. Removal percentage for lead by a biofilm of *Arthrobacter viscosus* supported on granular activated carbon, with surface treated with HNO_3 and H_2O_2. The initial concentration of lead is 8 mg/l (a) and 17 mg/l (b), with a flow rate of 10 ml min^{-1} and a residence time of 1.2 min.

Effect of the surface treatment of carbons

The surface treatment seems to have influence on the removal percentage of lead and iron (Figure 3, Figure 4 and Figure 5). The best results obtained with GAC treated with HNO_3 seems to be connected with the higher presence of acidic groups in the surface of this kind of GAC. This acidic group contributes to a better adhesion of the biofilm and consequently to a higher removal (Avery and Tobin, 1992). The better adhesion of the biofilm to the GAC-HNO_3 is

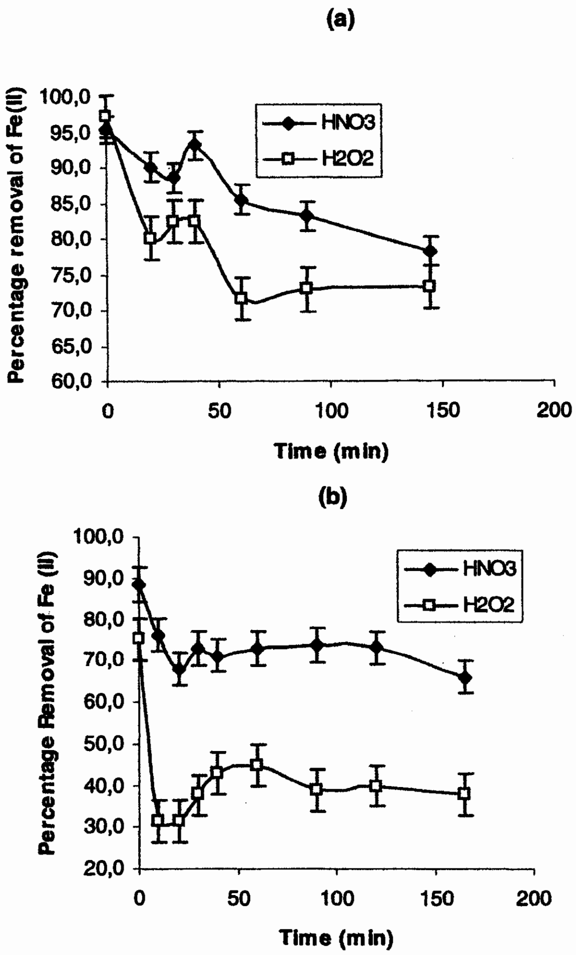


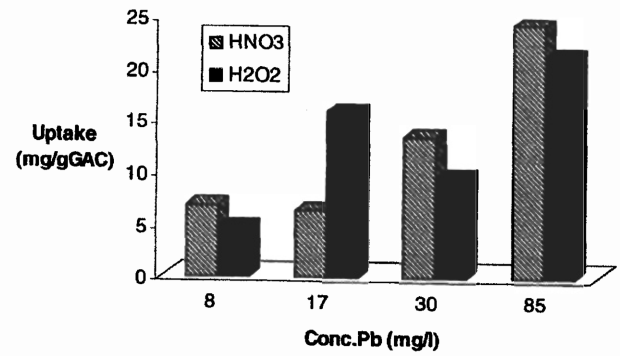
Figure 4. Removal percentage for iron by a biofilm of *Arthrobacter viscosus* supported on granular activated carbon, with surface treated with HNO_3 and H_2O_2. The initial concentration of lead is 5 mg/l (a) and 10 mg/l (b), with a flow rate of 10 ml min^{-1} and a residence time of 1.2 min.

confirmed by the higher production of polysaccharides and total polymers (Figure 2).

Uptake values

Uptake increases with the increase of the initial concentration of metals, as theoretically expected (Figure 5), and presents best results with GAC treated

(a)



(b)

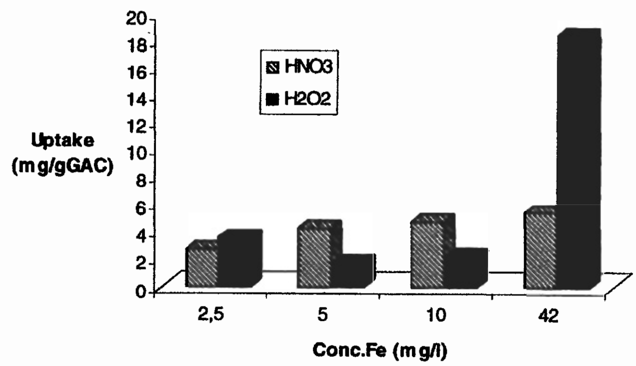


Figure 5. Final uptake values for lead (a) and iron (b) and for both GAC types for all the initial concentration tested.

with HNO_3. The final uptakes ranged from 4.8 mg_{Pb}/g_{GAC} to 24.2 mg_{Pb}/g_{GAC} and 1.8 mg_{Fe}/g_{GAC} to 18.4 mg_{Fe}/g_{GAC}. The results indicated that the range of concentrations used does not allow the saturation of the system. Studies with higher concentrations are being developed at this moment. The uptake value obtained for an initial concentration of iron of 42 mg l^{-1} and with GAC-H_2O_2 probably is affected by impurities on the biosorption system.

CONCLUSIONS

A biofilm of *Arthrobacter viscosus* supported on granular activated carbon is able to remove lead and iron from dilute solutions and can be applied in wastewater remediation. The best results were obtained during the first hour and for the most diluted solutions. The good results obtained with this system seem to be connected with the affinity between the cationic charge of the metals and the anionic charge of the bacteria, the elevated atomic weight of lead and the reduced ionic radius of iron. The production of polysaccharide and the biosorbent retention ability seems to be intimately related, that is, the optimization of the support surface is of great importance on the retention improvement.

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