

# Bacterial biofilm supported on granular activated carbon and on natural zeolites- an application to wastewater treatment

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## Introduction

The removal of many heavy metals from industrial wastewater is one of the most important environmental problems to be solved today. The retention of this contaminants by a biofilm supported on granular activated carbon or on natural zeolites is one of the promising technologies for the reduction of this problem, because it is cheap and it removes a broad range of substances, heavy metals and organic compounds.

This study aims the development of a system of two mini-columns in series for the removal of chromium (VI) using a biofilm of *Arthrobacter viscosus* supported on two different materials: granular activated carbon and natural zeolite. The effect of the regeneration of granular activated carbon was also studied.

## Material and Methods

### Materials

The bacterium *Arthrobacter viscosus* was obtained from the Spanish Type Culture Collection of the University of Valência. Aqueous chromium solutions were prepared by diluting  $K_2Cr_2O_7$  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 interference by other metals. Atomic absorption spectrometric standards were prepared from  $1000\text{ mg}_{Cr}\text{ l}^{-1}$  solution.

The supports were granular activated carbon (GAC) from MERCK with an average particle size of 2.5 mm, characterised by  $N_2$  adsorption (77K) with an ASAP Micromeritics 2001 which indicated a Langmuir area of  $1270\text{ m}^2\text{ g}^{-1}$  and an average pore diameter of 2 nm, and naturally occurring zeolite from Cuba, mainly composed by clinoptilolite,  $Al_2O_3 \cdot SiO_2$ , with a pore diameter between 0.2 and 1.2 nm and a internal surface smaller than  $10^3\text{ m}^2 \cdot \text{g}^{-1}$ . This last support was randomly smashed and only particles with a size between 2 and 5mm were used. Any attempt to reduce more the size of these particles would reduce to powder an important amount of the material. The option for granular supports is justified by the fact that this kind of biofilm demands detached particles that allow the bed to be expanded, avoiding the gluing by the exopolysaccharides produced by the bacteria. The selection of GAC was due to its high surface area, porous structure, high adsorption capacity and surface chemical nature, making it a versatile adsorbent and the zeolite was chosen due to its capacity for immobilising microorganisms and their ability to remove heavy metals from industrial wastewater.

### Methods

The whole experimental work was conducted in duplicate. GAC and the zeolite were placed separately, in Erlenmeyer flasks of 250 ml with 150 ml of distilled water. They were sterilised at  $120^{\circ}\text{C}$  for 20 min to release the air inside the pores. Then, those materials were placed in mini-columns (internal diameter = 0.9 cm, ht = 30 cm) for open system studies.

Three different set-up's were used: two columns in series filled with granular activated carbon, two columns in series filled with zeolite and a column with GAC (23 g) followed by another column with zeolite (40 g). The microorganism culture and the nutrient broth were pumped through at a flow rate of 25 ml/min, firstly to a mixture reactor and afterwards through the sequential beds, aiming the formation of the biofilm. Two different media, with different concentration of peptone, were used to grow the microorganism for 4 d, aiming the optimisation of the adhesion. The high flow rate used allows the formation of a compact biofilm and consequently a resistant one to the erosion stress resultant from the hydrodynamic forces.

After the biofilm formation, the beds were washed out and the metal solution, with controlled pH, was passed through the columns with a flow rate of 10 ml/min. At the end of each run, columns were washed out and samples of the effluent were seeded in Petri plates with nutrient agar to assess the metabolic activity of the microorganism. The GAC was recovered with steam and reused twice. Cr (VI) concentration at the inlet and at the outlet of the sequential columns was measured by Atomic Absorption Spectroscopy, Varian Spectra AA-250 Plus.

## Results and Discussion

The biosorption of Cr(VI) was considered and Figure 1 shows the dependence of the total uptake of the metallic ion as a function of the initial concentration on the water solution. There is a clear maximum around 70 mg.l<sup>-1</sup> and this value established the initial conditions of the future experiments. This maximum is probably defined by the balance between the effect of the increasing driving force in terms of concentration difference between the bulk solution and the biosorbent surface and the effect of the saturation of the surface matrix.

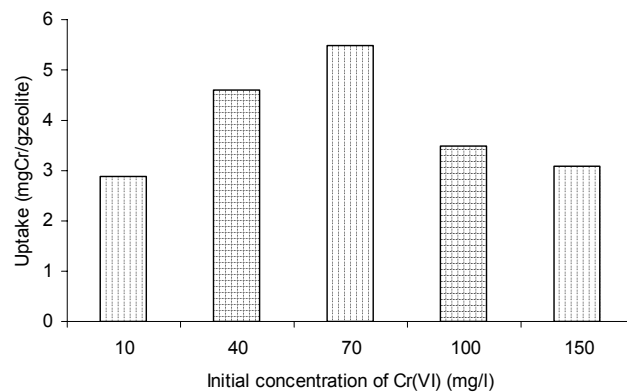


Figure 1. Uptake of Cr (VI) by a biofilm of *Arthrobacter viscosus* supported on a zeolite as a function of the initial concentration of the metal in the water solution to be treated.

The zeolite is not as good adsorbent as GAC, as its open cage structure is not so sensitive to the biofilm effect. The combination of the two supports shows a similar trend to the one of the two GAC beds, more accentuated, as residual metal ions in bulk solution will be entrapped easier in the second bed. After 27 h of continuous treatment, the removal of Cr (VI) by two beds of zeolite reaches 18%. Similar value is reached with two beds of GAC, 19%. It increases to 42% when both supports were used in series connection, with an initial concentration of 70 mg<sub>Cr</sub>l<sup>-1</sup> (Figure 2).

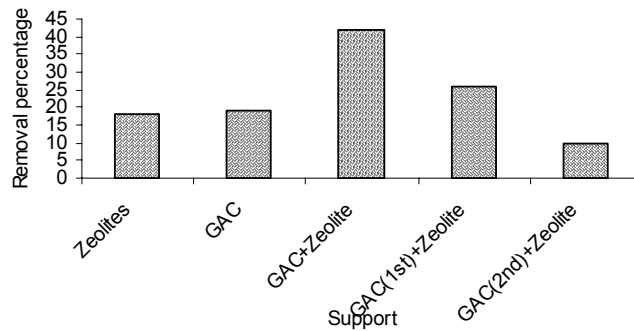


Figure 2- Removal percentage values for the three different supports after 27 hours. The initial concentration of chromium was  $70 \text{ mg l}^{-1}$ , with a flow rate of  $10 \text{ ml.min}^{-1}$ .

The regeneration of the support is important to reduce the high costs associated to the activated carbon. The steaming of GAC reduced the removal capability of the system in 38% and 76%, respectively for the 1<sup>st</sup> and 2<sup>nd</sup> regeneration (Figure 3).

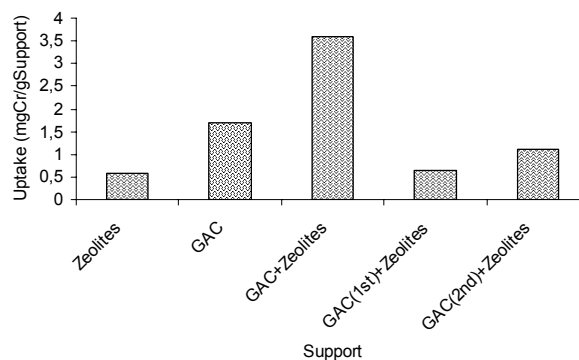


Figure 3. Uptake values for two beds of zeolite support, two beds of GAC support, mixed system, mixed system after 1 regeneration of GAC bed and mixed system after 2 regenerations of GAC bed.

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