Removal of hexavalent chromium from waste water using immobilized *Bacillus brevis*

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Abstract

Conventional methods used to remove chromium(VI)) from waste water include ion exchange or adsorption on activated carbon. A viable alternative for removal of metals may be the application of biosorption. In this work, chromium (VI) removal is studied in a batch system using immobilized Bacillus brevis beads. The effect of initial concentrations of chromium and itspercentage removal is studied by conducting experiments at different initial concentrations ranging from 10 to 50 mg/l.

The percentage removal is higher in the initial stages. The impact of the Cr(VI) uptake is studied for various initial concentrations like 20,30,50 mg/l and it is found that the Cr(VI) uptake is reduced for high pH values.

Keywords: Hexavalent Chromium, *Bacillus brevis*, Biosorption.

Introduction

The development of industry has an enormous effect on the improvement of life quality of human being^{1,2}. The accelerated industrial activities have exaggerated the environmental pollution and deteriorate the natural ecosystem. It causes the accumulation of pollutants such as heavy metals, synthetic compounds, waste nuclear liquids etc.^{3,4}

Constantly paying attention to the potential health hazard is presented by heavy metals to the environment⁵⁻⁷. There are two predominant forms of Cr namely trivalent and hexavalent which are found in industrial waste waters. Some of major industries discharging Cr (VI) in their effluents are chrome plating, textile, leather tanning, electro plating, pigment and dyes, metallurgical, metal finishing, photography and wood preserving.

Small amounts are used in drilling muds, rusts and corrosion inhibitors and toner for copying machine⁸. Human health is affected when Cr (VI) is exposed in excess of prescribed level. Cr (VI) is well known for its toxicity. Concentrations of Cr (VI) greater than 2 μ g can cause irritation to nose, sneezing, itching etc. It can generate disorders and diseases when accumulated at high levels and it can ultimately become lethal. Some Cr (VI) compounds like calcium chromate, lead chromate, Zinc chromate etc. are known as carcinogens. Mining and metallurgical waste waters are considered to be the major sources of heavy metal contamination^{9,10}.

Material and Methods

Micro Organism and Growth Conditions: Bacterium called *Bacillus brevis* was selected for this study which is collected at (MTCC) Microbial Type culture collection center of the Institute of Microbial Technology (IMTECH), Chandigarh, India based on its ability to remove heavy metals and the chemicals used were of the highest pure grade commercially available.

Procedure for Maintenance of Cells: The culture *Bacillus brevis* are maintained in the agar media. Peptone 0.5%, meat extract 0.3%, sodium chloride 0.5%, adjusting pH from 7.2 - 7.4 with sodium hydroxide solution and 0.3% agar were used for the preparation of slants.

For growing the media on a large scale, fresh culture slants were transferred to 100ml of liquid media containing peptone 0.5%, meat extract 0.3%, sodium chloride 0.5%, pH 7.2-7.4. The media was left 2 days for growth. 100ml media was transferred into 500ml media and the culture was used for further studies. Here in all the cases, the media was autoclaved under 121°C at 15psi for 20 minutes and strict aseptic conditions were maintained throughout the experiment by taking precautions while inoculating and transforming the culture.

Preparation of Immobilized Cells: For immobilization of *Bacillus brevis* cell, sodium alginate was used as a matrix. The cells were transferred aseptically to centrifuge tube and centrifuged for 5 minutes, the supernatant liquid was decanted and the cell past was suspended in sodium alginate solution. *Bacillus brevis* - alginate suspension was made by adding 10gm of cells along with 8% of sodium alginate. The resulting slurry was extruded as drops into 2% calcium chloride solution at room temperature and maintains strict aseptic conditions while preparing the immobilized cells. The beads were hardened and the size of the beads could be varied.

Preparation of Cr (VI) Solution: A test solution containing Cr was made by diluting 1ml of stock solution of metal to the desired concentrations. The ranges of concentrations of Cr prepared from stock solutions varied between 10 to 50 mg/l. A stock solution of synthetic wastes Cr (VI) was obtained by dissolving same volume of potassium dichromate in double-distilled and deionized water.

Biosorption Experiments: Batch mode biosorption studies were conducted by adding 1gm of bio sorbent in 100ml of a synthetic Cr (VI) solution at desired concentrations. These were agitated with pre-determined time intervals; the bio sorbent was separated and the supernatant liquid was analyzed spectrophotometrically for the remaining Cr (VI) concentrations. The effect of initial Cr (VI) concentrations at different agitation time was carried out.

For pH studies, 100ml of different concentrations of Cr (VI) solutions (20, 30, 50 mg/l) were adjusted to different pH values (2, 5, 6, 7) agitated. Spectrophotometer is used to analyze the remaining Cr (VI) solution.

Results and Discussion

Effect of Initial Concentration in Chromium (VI) Uptake: The effect of Cr (VI) uptake and percent removal of Cr (VI) with time for different concentrations was noticed. From fig. 1, it is observed that the Cr(VI) uptake on biosorbent increases both by increase in contact time and initial concentration of Cr (VI) attains equilibrium after 24 hours.

From fig. 2 and 3, It is analyzed that the percent removal of Cr (VI) increases with increase in contact time and decreases with increasing initial concentration of Cr (VI). The percentage removal of Cr (VI) is higher in the initial stages because in these stages, adequate surface area of bio sorbent is available for biosorption. Chromiumr (VI) on percentage removal was found to be 56%, 36%, 23%, 21% and 17% at the initial Cr concentrations of 10 to 50mg/l

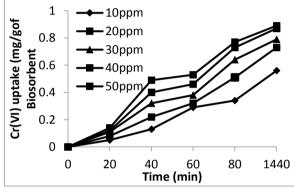


Fig. 1: Effect of Contact Time on Cr (IV) Uptake for various initial concentrations

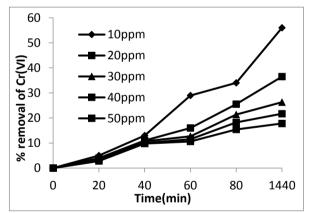


Fig. 2: Effect of Contact Time on % removal of Cr (VI)

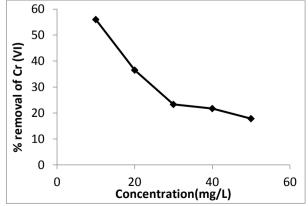


Fig. 3: Effect of Concentration on Percentage removal of Chromium (VI) at equilibrium condition

Effect of pH on Cr (VI) uptake and percentage removal of Cr (VI): The effect of pH on the Cr (VI) uptake and percentage removal of Cr (VI) for various initial concentration (20,30,50 mg/l) was noticed. From figures 4 to 7, it is observed that the Cr (VI) uptake in bio sorbent increases with increase in contact time and decreases with increase in pH. The lower pH values also yielded increased uptake of anionic Cr (VI). The absorbent in aqueous solution has surface charges on ionizable functional groups. The surface charge is dependent on the types of compounds in cell walls of the biomass mainly responsible for biosorption. The cell walls of gram-positive bacteria are mainly composed peptidoglycan, proteins and amino acids.

As the pH of the solution decreased, negatively charged carboxyl groups and neutral weak base amine groups on biomass became protonated, offering positive binding sites for Cr (VI). With increase in pH of the system, the degree of protonation of the surface reduced gradually and hence decreased Cr (VI) uptake was noticed.

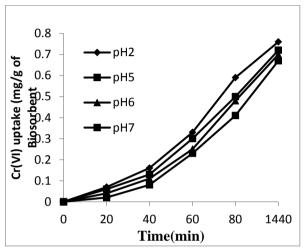


Fig. 4: Effect of contact Time on Cr (VI) uptake of 20 mg/l

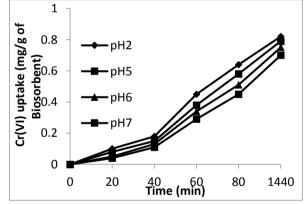


Fig. 5: Effect of contact Time on Cr (VI) Uptake of 30 mg/l

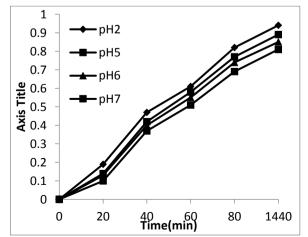


Fig. 6: Effect of contact Time on Cr (VI) uptake of 50 mg/l

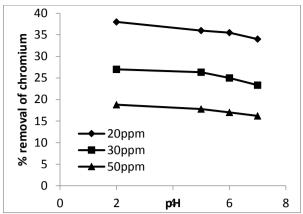


Fig. 7: Effect of pH on Percentage removal of Cr (VI) for various concentrations at equilibrium

Conclusion

The entrapment technique was adapted for the whole cell immobilization method. The percent removal of chromium (VI) was higher in the initial stages and decreased with increasing initial concentration of chromium. With the increase in the pH of the system, the degree of protonation of the surface reduces gradually and hence chromium (VI) uptake decreased.

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