Microbial Bioremediation of Chromium and Development of Enzymatic Biosensor by Enterobacter Aerogenes T2 (GU265554; NII 1111)

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Abstract

Carcinogenic heavy metal chromium, in its hexavalent form (Cr(VI)), is spuriously used in various industrial operations because of its hardness and stability and is found in industrial effluents in concentrations much above WHO’s prescribed limit (50 mg/L). Conventional methods for removing toxic chromium ion (by chemical reduction followed by precipitation, ion-exchange and adsorption on activated coal, alum, kaolinite and ash) are costly for large scale treatment. Microbial uptake followed by reduction of toxic Cr(VI) may be very successful since biological strategies provide cost effective green technology. In this study, we have isolated a new powerful chromium resistant bacterium isolated from tannery waste and studied its utilization in bioremediation as well as biosensing of Cr(VI).

1. Introduction

Cr (VI) which enters the environment through several anthropogenic activities is highly toxic and carcinogenic. Several health hazards are associated with Cr(VI) exposure in humans, including lung carcinoma and impaired foetal development in mammal (Srinath et al, 2002). Hence, detection or sensing along with quantification and subsequent remediation of Cr(VI) (to non-toxic trivalent chromium) from industrial effluent, drinking water is necessary. In aqueous systems Cr(VI) exists as oxyanions which are structurally analogous to sulphate ions, hence can easily be taken up by anionic transport systems of bacterial cells. Their cell-membrane is nearly impermeable to Cr(III) and thus Cr(III) has one-thousandth of the toxicity of Cr(VI).
Because of the insolubility, Cr(III) facilitates its precipitation and removal, and hence the biotransformation of Cr(VI) to Cr(III) has been considered as an alternative process for treating Cr(VI)-contaminated wastes.

2. Methodologies and Results
2.1 Identification of most efficient strain and its use in bioremediation and biosensing
This strain was first isolated from tannery effluents on selective media plates, characterized through various methods including biochemical tests, antibiotic assay, 16SrRNA gene sequencing etc. and was identified as *Enterobacter aerogenes* T2 GU265554; NII 1111 (Panda and Sarkar, 2012). It performed very well by removing about 99% (average) from a synthetic culture media having 20 mg/L Cr(VI). *Enterobacter aerogenes* T2 was later used successfully to remediate Cr(VI) from soil (Fig. 1), of potted plants. T2 was used to remediate chromium from tannery effluents in a laboratory level experiment.

![Figure 1](image.png)

*Figure 1*: Plant B was treated with 5 ppm Cr(VI) solution along with T2 bacterial suspension and the plant was visibly unaffected after 7 days, whereas leaves of plant A were affected.

Hence, further bioremediation experiments were conducted by first immobilizing the cell free extract into calcium alginate beads and then observing the removal of Cr(VI) by batch (Fig. 2).
Figure 2: Bioremediation of 10 mg/L Cr(VI) by alginate-T2 beads; (Inset) SEM photograph of T2 bacteria, resolution = 10 μm.

2.2 Microbe–metal interaction study
Microbe-metal interactions (Beveridge et al, 2000) were further studied by various Electron Microscopies (Fig. 3) and chromium peak was observed in Energy Dispersive X-ray Spectroscopic microanalysis (Panda and Sarkar, 2012). This investigation revealed that Enterobacter aerogenes T2 helped remediate a moderate amount of Cr(VI) (8–16 mg/L) over a wide range of pH values at 35–37°C (within 26.05 h).

Figure 3: Scanning Electron Microscopic image of fresh whole alginate-T2 beads; Resolution = 500μm; Energy Dispersive X-ray Spectroscopic microanalysis of used beads showing Cr(VI) accumulation.
Crude cell free extract (CFE) of Enterobacter aerogenes T2 was exploited to develop a stable biosensor for direct estimation of Cr(VI) in waste water, by using three-electrode assembly via cyclic voltammetry (Fig. 4). The proposed sensor showed linear response in the range of 10-40 µg/L Cr(VI) and the limit of detection was found to be 6.6 µg/L Cr(VI). No interference was observed in presence of other metal ions.

![Figure 4: Cyclic voltamogram of various concentrations of Cr (VI) (scan rate= 10mV/s, Potential = -1.0 to +0.3 V).](image)

3. Conclusion
The proposed sensing system could be a viable alternative to costly measurement procedures. Calcium alginate beads, modified with CFE of Enterobacter aerogenes T2, could be used in bioremediation of Cr(VI) since it could work in real conditions with extraordinarily high capacity.

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References


