Accumulation of Cr(VI) Reducing Bacteria by Electrochemical Cultivation

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Introduction

We have obtained chromium reducing bacteria from soil. These bacteria not only thrive at concentrations of toxic hexavalent chromium, Cr(VI), ions that are toxic to most bacteria, but under anaerobic conditions, also reduce Cr(VI) to insoluble Cr(III). However, obtaining a pure culture of this bacteria using conventional cultivation condition is difficult because as the concentration of Cr(VI) ions decreases, dominant bacteria from the original inoculation begin to thrive. Here we present results obtained using an electrochemical cell to replenish the Cr(VI) ions thus maintaining an environment not conducive to the growth of the dominant bacteria.

Experimental

Cr(VI) reducing bacteria was collected from an environmental soil sample and cultivated for 1 month under anaerobic condition in a medium with the following contents (g/L); NH_4Cl, 0.132; K_2HPO_4, 0.041; $MgCl_2$ $6H_2O$, 0.49; $CaCl_2$ $2H_2O$, 0.009; KCl, 0.052; $K_2Cr_2O_7$, 0.015, sodium lactate, 1.0 and Yeast extract, 0.2. After 1 month of cultivation, Cr(VI) in the medium was completely reduced to Cr(III) and several species of bacteria were evident in the culture.

An electrochemical cultivation cell was constructed, consisting of a Pt electrode (anode) and a carbon electrode (cathode) separated by an ion exchange membrane as shown in Figure 1. For electrochemical cultivations, 10 mL of the bacteria mixture and 90 mL of fresh medium was inoculated into the anode well in each of three cultivation cells. One hundred mL of Cr(VI) free medium was poured in each cathode. All three baths were set in an anaerobic box and the potentials described below were applied. After 10 days cultivation at 30 °C, the cell growth for each bath was counted. In addition, the physical characteristics of the cultured bacteria were examined using atomic force microscopy (AFM).

Results and Discussion

A cyclic voltammogram for Cr in solution is shown in Figure 2. Reduction and oxidation of Cr occurred at 0.1 V and 1.0 V (vs. Ag/AgCl), respectively. Based on this result, the potentials of the electrochemical cultivation cells were set at 0.2V, 0.6V and 1.0V (vs. Ag/AgCl). After 10 days incubation, cell density in the 0.2 V and 0.6 V cells was about 4 x 10^7 cells/mL, that was almost the same as initial value, while, about 33% higher than the initial (Fig 3). Cell growth in the 1.0 V cell was over 8 x 10^7 cells/mL, about 166% higher than the initial. The potential inducing growth enhancement (1.0V) is well matched to that of Cr(VI) regeneration shown in Fig 2. This result suggests the Cr(VI) reducing bacteria requires Cr(VI) ions for growth.

Under microscopic and AFM examination, bacteria grown by electrochemical cultivation were nearly uniform in appearance (Fig 4b), in contrast to the mixture apparent in the initial solution (Fig 4a). Our results show clearly that growth of Cr(VI) reducing bacteria is enhanced by continuously supplying Cr(VI) ions through electrochemical cultivation.

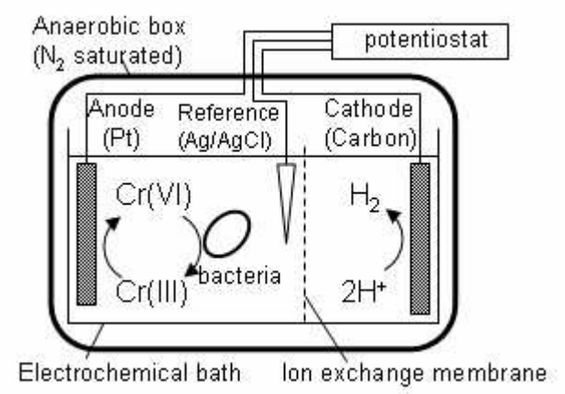

Figure 1. Electrochemical cultivation system.

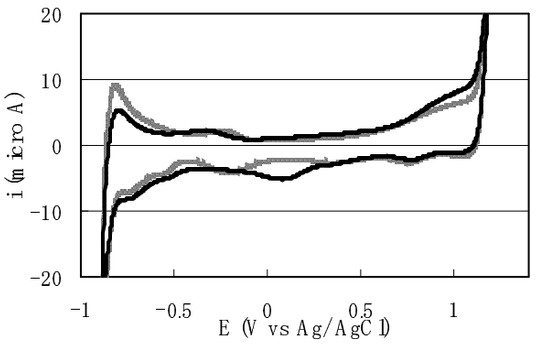

Figure 2. Cycric voltammogram of 10 mM of Cr ions in KCl. Working electrode: Pt; scan rate: 100 mV/s. Gray line represented baseline.

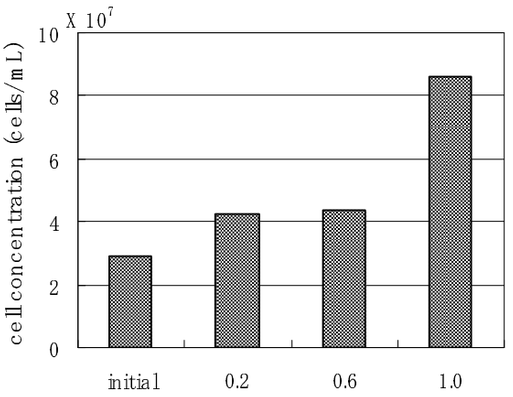

Figure 3. Growth of bacteria after 10 days electrochemical cultivation with varied potential.

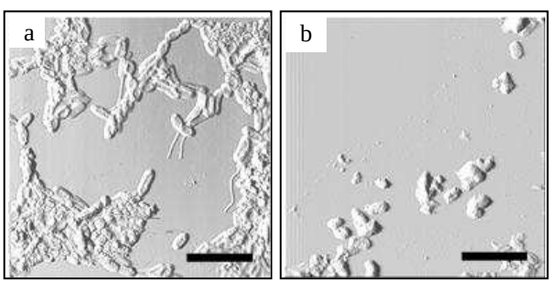

Figure 4. AFM image of bacteria from initial solution (a) and from after 10 days electrochemical cultivation with the potential of 1.0 V (b). Bars; 5μm.