EFFECTS OF pH AND IONIC STRENGTH ON THE ADSORPTION OF Cs, Sr, Eu, Zn, Cd AND Hg BY PSEUDOMONAS PUTIDA

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Abstract. Bacterial metal accumulation may influence the mobility and chemical form (speciation) of metals in the environment. The passive adsorption of six metals (Cs, Sr, Eu, Zn, Cd and Hg) by a soil bacterium, Pseudomonas putida, was studied in the present work, using a radiotracer batch-distribution technique. To replicate natural conditions, the adsorption was considered as a function of pH (4–10) and ionic strength (0.01 M and 0.1 M KCl) at a low metal concentration (10–8 M). P. putida exhibited a total metal accumulating capacity of 200–1000 meq kg–1 bacteria (dry weight) (measured in 0.01 M KCl at pH 6.4). This capacity is comparable to that of many organic soil components and it is above the capacity of most inorganic constituents. The following affinity order of adsorption was observed: Hg > Eu > Cd, Zn, Sr > Cs. The results indicate that the bacterial surface carries different sites that exhibit varying affinity and capacity for binding metal ions. It can be concluded that the overall adsorption of metals by P. putida is determined by several interacting processes related to the properties of both the metals and the bacterial surface and to the composition of the solution phase (pH as well as ionic strength).

Key words: bacteria, metals, accumulation, metal mobility

1. Introduction

The dispersion of metals in the environment has increased as a result of anthropogenic activities and has, in particular, been accelerated by the progressing regional soil acidification. This emphasizes the importance of gaining detailed knowledge regarding the mechanisms that regulate the mobility of metals in soil and water and the role of the various soil components, including microorganisms, in the distribution of metals in the soil.

 Constituents of cell walls of both Gram-positive and Gram-negative bacteria have been found to interact strongly with metal ions (for review, see Beveridge, 1986). Such metal accumulation can affect the transport properties of trace metals and their overall distribution in the ecosystem (Ford and Mitchell, 1992). The objective of many studies of bacteria-metal interaction has been to find species that can be suitable either to detoxify polluted waters or recover precious metals, i.e. that have high selectivity or total accumulation capacity (for review, see Gadd, 1988). The experimental conditions applied in those studies were more or less artificial in terms of, among other parameters, metal concentration and pH, and

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few studies have dealt with adsorption phenomena at metal concentrations representative of natural background levels. Consequently, the importance of bacterial metal accumulation under typical environmental conditions is poorly understood. The importance of pH for bacterial metal accumulation has been demonstrated (Pedersen and Albinsson, 1991; Premuzic et al., 1991; Wallberg et al., 1991). On the other hand, there is very little, if any, data concerning the influence of ionic strength, particularly the combined effects of pH and ionic strength on bacterial metal adsorption.

The aim of the present investigation was to determine to what extent some trace metals are accumulated by a common soil bacterium, *Pseudomonas putida*, at metal concentration levels representative of unpolluted natural soil-groundwater systems. The influence of pH and ionic strength on the adsorption was also considered. Three metals, i.e. Cs, Sr and Eu, representing the oxidation states I, II and III were selected. In addition, three heavy metals, i.e. Zn, Cd and Hg, all from the IIB group, were chosen because they are of environmental concern.

2. Experimental

2.1. Bacteria and Growth Conditions

*Pseudomonas putida* (CCUG 28920) isolated from soil was grown at 23 °C in a citrate medium (pH 6.0) with a composition described elsewhere (Brynildsen et al., 1988). The culture was harvested at late log phase, washed and starved in 0.1 or 0.01 M KCl as previously described (Wallberg et al., 1991). Cell density was determined from the optical density (A550) correlated to dry weight and viable counts.

2.2. Metal Adsorption Studies

Metal distribution studies were performed in KCl solution at two ionic strengths (0.01 and 0.1 M), using a radiotracer batch distribution technique (Wallberg et al., 1991). The nuclides used were $^{134}$Cs, $^{85}$Sr, $^{152}$Eu, $^{65}$Zn, $^{109}$Cd and $^{203}$Hg, with addition of non-radioactive carriers of the same metals to obtain the desired total metal concentration (usually $10^{-8}$ M). Studies were performed in 50-ml polyallomer centrifuge tubes (Nalgene) with a total sample volume of 20 ml. The various metals were added as chlorides, and the pH was adjusted by adding HCl or KOH (filter sterilized) to obtain values ranging from 3 to 10. No buffer was added for pH control; consequently very few of the systems (particularly between pH 7 and 9) reached a final pH that was in the near-neutral range. Starved bacteria were added to yield a density of 0.075 mg (dry weight) per ml ($2.4 \times 10^8$ CFU ml$^{-1}$). The tubes were placed on a rotary shaker (120 rpm) at room temperature for 24 h and were thereafter centrifuged. The supernatants were analyzed for metal content by using a well-type gamma counter (Compu Gamma, LKB), and the pH