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Potential for biocolloid transport of cesium at high ionic strength

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HIGHLIGHTS

- No *Chromohalobacter* interaction with cesium at high salt.
- No adsorption and intracellular uptake of cesium with *Chromohalobacter*.
- Interaction of cesium with dolomite at high ionic strength.

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ABSTRACT

In subsurface repositories, active bacterial populations may directly influence the fate and transport of radionuclides including in salt repository systems like the Waste Isolation Pilot Plant in Carlsbad, NM. This research quantified the potential for transport and interaction between Chromohalobacter sp. and Cs in a high ionic strength system (2.6 M NaCl) containing natural minerals. Mini-column experiments showed that Chromohalobacter moved nearly un-retarded under these conditions and that there was neither association of Cs with microbes nor dolomite despite changes in bacterial metabolic phases. Growth batch experiments that monitored the potential uptake of Cs into the microbes confirmed results in column experiments where intracellular uptake of Cs by Chromohalobacter was not observed. These results show that Cs may be highly mobile if released in high ionic strength systems and/or carbonate minerals with negligible inhibition by these microbes.

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1. Introduction

Radioactive cesium (Cs) is produced during nuclear fission of uranium-235 with a yield of approximately 6% for ^{137}Cs (half-life 30.2 years). Approximately 6.8×10^7 Ci of ^{137}Cs are produced annually in the U.S. primarily in commercial nuclear power reactors (Okumura, 2003) and require proper management and disposal. Previous releases have occurred internationally to the environment including high ionic strength systems. These environmental releases may impact future drinking water sources as well as other exposure vectors. For example, significant atmospheric releases and deposition to soils and seawater occurred at the Fukushima Daiichi Nuclear Power Plant (FDNPP) in 2011 with an estimated 1.5×10^5 Ci (Ci) of ^{137}Cs deposited on the Japanese islands and the surrounding ocean (Kubo et al., 2018; Yasunari et al., 2011).

Proper disposal of radioactive waste is vital to decrease the future risk of exposure to the public. Long term radioactive waste disposal is ongoing within geological formations including at the Waste Isolation Pilot Plant (WIPP). The WIPP is a deep geologic repository located in a salt formation outside of Carlsbad, NM which is expected have an inventory of 2.5×10^5 Ci of ^{137}Cs by 2033 based on the amount of waste currently remaining at generator sites throughout the U.S. and awaiting transport to the WIPP (Patterson and Shrader, 2018; Van Soest et al., 2018). This facility is located in a salt bed with high ionic strength brines, further highlighting the need to quantify potential transport phenomena for radionuclides for modeling and risk assessment activities.

Cs fate may be affected by different factors in environmental

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Abbreviations

HPW – high purity water, > 18 MΩ*cm
ICP-MS – inductively coupled plasma mass spectrometer
MWCO – molecular weight cut-off
NPT – National Pipe Taper
PEEK – polyetheretherketone
PTFE – polytetrafluoroethylene
TEM – transmission electron microscope
WIPP – Waste Isolation Pilot Plant

systems including mineral phases, microbes, and salt content. Previous research at low ionic strength has shown that Cs can accumulate with mineral phases and reacts primarily through ion exchange (Goto et al., 2014; Montgomery et al., 2017; Zaunbrecher et al., 2015). For example, Montgomery and team reported that Cs exhibited weak sorption to Savannah River Site minerals as the total Cs concentration increased. Therefore, Cs may be highly mobile in high ionic strength systems due to competition with similar cations, as it is dominant as a monovalent cation (Montgomery et al., 2017; Saiers and Hornberger, 1999).

Microbes also have been shown to affect the fate of radionuclides including ^{137}Cs with the potential for both surface adsorption and intracellular uptake (Francis, 2012; Gadd, 2010; Zhong et al., 2017). Microorganisms have active roles in the environment and their properties can affect metal speciation, toxicity, mobility, and mineral dissolution (Francis, 2012; Gadd, 2010). Further, contaminants and minerals in both anthropogenic and natural environments can impact the growth, activity, and survival potential of microbial populations, although the impact of microorganisms under high salt conditions on Cs mobility is not yet fully understood.

Research has shown that Cs can accumulate in the biological food chain especially in place of potassium due to their similar chemistry (Avery, 1996). The potential for intracellular uptake of Cs into microbes has been demonstrated at low ionic strength (Bossemeyer et al., 1989; Johnson et al., 1991; Ohnuki et al., 2003; Tomioka et al., 1992, 1994). For example, Tomioka reported accumulation of as much as 92 mg/g of Cs by dry cell weight in *Rhodococcus erythropolis* CS98 (Tomioka et al., 1994). Bossemeyer also showed that Cs can be taken up by *Escherichia coli* at low ionic strength through the same mechanism as K uptake with K-replete cells taking up Cs proportionally to the ratio of Cs and K in the aqueous phase (Bossemeyer et al., 1989). However, previous research has not considered the potential for Cs uptake at high ionic strength.

Researchers previously observed an increase in biosorption of Np, Cm, and Eu with increased ionic strength onto the surface of halophilic bacteria (Ams et al., 2013; Takenaka et al., 2004). These researchers suggested that the increase in adsorption with ionic strength may be attributed to increased specific surface area and hydrophilicity of the cells (Takenaka et al., 2004) or ion activities (Ams et al., 2013). These results highlight the potential for cation adsorption to halophilic microbes at high ionic strength and the need to better understand their complex interactions.

There is a need to understand the potential for adsorption to and intracellular uptake of Cs by microbes in high ionic strength systems. The objective of this research is to quantify (1) the potential for transport of the halophilic bacterium *Chromohalobacter* and (2) the influence of these microbes on the mobilization of Cs in high ionic strength systems. This study focused on conditions relevant to the WIPP to apply this research to a real system.

2. Materials and methods

2.1. Mineral collection and characterization

Dolomite [$CaMg(CO_3)_2$] rock samples were collected from the Culebra bluff outcrop located near the Pecos River parallel to the WIPP (Emerson et al., 2018). The rock samples were stored in an airtight, plastic container at room temperature prior to experiments. The rocks were crushed with a stainless-steel impact mortar and pestle (catalogue no. 850, Chemplex, Palm City, FL; USA), sieved (No. 45, 100 and 200 sizes, Fisher, Stainless steel), cleansed with high purity water (HPW, >18 MΩ*cm) and finally placed in an oven at 40 °C for drying for at least 18 h (Emerson et al., 2018). Culebra Dolomite utilized in these experiments ranged from 355 to 500 μm in diameter to allow sufficient surface area for interaction with large enough particles to avoid clogging in columns. The washing step with HPW was conducted to cleanse and remove surface impurities from the dolomite. The process was repeated three times prior to use in experiments. Bulk surface area was measured via the Brunauer-Emmett-Teller (BET) method (Micromeritics TriStar II 3020, Norcross, GA; USA) at 1.70 m^2/g. XRD and SEM-EDS was conducted previously to confirm nearly 99% dolomite (Emerson et al., 2018). The XRD and SEM-EDS results confirmed that the cleaning process removed surface impurities from nearby geologic layers during collection of the sample.

2.2. Chromohalobacter sp. strain PZ13 preparation and characterization

The halophilic bacterium, *Chromohalobacter* sp. strain PZ13, was originally isolated from the far-field groundwater of the WIPP and grown for these experiments. Fig. S1 of the *Supplemental Materials* contains an image of the bacteria using transmission electron microscopy. *Chromohalobacter* was grown in a generic halophile broth (GHB) containing 15% NaCl (w/v; equivalent to 2.6 M NaCl) and 3 mM bicarbonate and other micronutrients for growth including yeast extract and casamino acids as described previously (Zengotita et al., 2017) and in *Supplementary Materials* Table S1. Cells were harvested and concentrated by centrifugation. The microbes were washed three times with sterile 15% NaCl +3 mM $NaHCO_3$ solution. After the final wash, they were re-suspended in the test solution to obtain an optical density (OD) of 0.23–0.26 at 600 nm (Genesys 20 spectrophotometer, Daly City, CA; USA) which corresponded to approximately 10^8 cells/ml as shown in Fig. S2 in the *Supplemental Materials*.

Additional stationary phase experiments were also conducted where *Chromohalobacter* was allowed to grow for 30 h to reach stationary phase. The *Chromohalobacter* suspension was spiked with 200 ppb of ^{133}Cs diluted in HPW from a commercial stock (1000 ppm in 2% HNO_3, High Purity Standards, Charleston, SC; USA) and equilibrated for 1 h prior to injection. ^{133}Cs was utilized as a stable, non-radioactive analogue and exhibits identical chemical behavior to the radioactive isotopes of Cs. Further, stationary phase microbes were subjected to the following conditions: (1) stressed and (2) normal. A portion of the total *Chromohalobacter* stock was placed into the oven at 70 °C to induce a stressed state which was measured as a ratio of 60% stressed and 40% dead as determined after staining with the LIVE/DEAD BacLight viability kit (ThermoFisher Scientific). The other half of the stock was harvested prior to oven treatment in the stationary phase to observe the differences in microbial transport under these extreme conditions.

In the log-growth experiments, the cells were harvested after 14–16 h of growth. Each suspension was spiked with 5000 ppb of Cs either 1 h prior to injection or grown in the presence of Cs then spiked additionally. These conditions were investigated to observe

the potential differences in uptake between actively metabolizing and non-metabolizing cells.

2.3. Miniature column setup

Miniature, saturated column experiments were conducted based on previous designs and used as a small-scale representation for potential release through dolomite collected from the Rustler formation located above the WIPP (Dittrich et al., 2016; Emerson et al., 2018). Packed-column experiments have previously been utilized as models for bacterial transport (Gargiulo et al., 2007; Walker et al., 2005a, 2005b).

Briefly, one inch of polytetrafluoroethylene (PTFE) tubing (3/8″ inner diameter, International Polymer Solutions, Irvine, CA; USA) was packed with 1 g of Culebra dolomite [355–500 μm]. The tubings were threaded with a 1/8" 27 National Pipe Taper (NPT) carbon pipe tap (Drillco, Baton Rouge, LA; USA). The fittings were sealed with silicone and the inlet and outlet of the columns was covered with 35 μm polyetheretherketone (PEEK) mesh (Spectrum labs, Irving, TX; USA) to reduce the potential for clogging within the tubing. The miniature columns had an average porosity of 0.45, as determined by mass. Columns, samples, and the fraction collector remained within a clear plastic container throughout experiments to reduce evaporation.

Attached to each mini-column were 30 cm inlet and 60 cm outlet tubings (PTFE #20 0.032" inner diameter Cole-Parmer, Vernon Hills, IL; USA) that were connected to 60 mL polypropylene syringes. The syringes were attached to a syringe pump (Kd Scientific Model 100 series, Hollison, MA; USA) operating at a 0.013 mL/min flow rate. This flow rate is similar to the maximum hydraulic conductivity measured previously in the Culebra dolomite formation (1×10^{-5} m/sec) (Corbet, 2000). Therefore, it represents a worst-case scenario with the shortest potential interaction time of Cs with dolomite and microbes.

Prior to the start of the transport experiments, the miniature columns were equilibrated with the 15% NaCl +3 mM $NaHCO_3$ synthetic brine overnight to reach an average pH of 8.3. This allowed for the solutions to equilibrate with carbonate in the atmosphere and dolomite. In addition, this pH is similar to that in the repository setting and to the optimal pH for Cs uptake as suggested previously (Chen et al., 2005). After full saturation and equilibration with synthetic brine, *Chromohalobacter* suspensions were injected into columns from the bottom to induce a uniform flow against gravity. Alongside the biocolloid transport experiments, a negative control column was conducted to investigate Cs interaction with dolomite in the absence of microbes (Table 1). Conditions were varied to include injection of (1) log growth versus stationary phases of microbes, (2) stressed versus healthy microbes, and (3) microbes grown in the presence and absence of Cs.

2.4. Sampling protocol

The column effluent was accumulated into a fraction collector (FC203B Gilson; Middleton, WI; USA) with 13 × 100 mm polystyrene tubes at variable collection times. The effluent was weighed to track the total volume and flow rate by mass. Samples were regularly removed for analysis via ICP-MS for Cs. In addition, each aliquot was analyzed for pH following a three point calibration procedure. The optical density was measured at 600 nm (Genesys 20 spectrophotometer, Daly City, CA; USA) to identify the concentrations of *Chromohalobacter* over time following vortexing (Fisher Scientific) for 30 s to re-suspend all microbes in effluent as described previously (Zengotita et al., 2017). We previously determined that the optical density at this wavelength correlated with aqueous microbe concentrations (*Supplementary Materials* Fig. S2). Other researchers have also used measurements of the optical density at 578 and 600 nm to correlate with the concentration of microbes in similar fashion (Bossemeyer et al., 1989; Ohnuki et al., 2003; Takenaka et al., 2004).

To measure Cs in the aqueous phase or the fraction not associated with the microbes, unfiltered and filtered samples were collected, respectively. From the effluent collection tubes, 20 μL of sample was transferred into 2% HNO_3, and the acidified sample was then stored for ~24 h to achieve total cell lysis. After the lysis period, the unfiltered sample was collected and diluted in 2% HNO_3 for ICP-MS analysis. The filtered samples did not require a lysis step because the filter removes all cells although they were also prepared in 2% HNO_3 for ICP-MS analysis. An aliquot of sample was filtered in 100 kD molecular weight cut-off (MWCO, Pall Omega Nanosep, Show Low, AZ; USA) filters with centrifugation for 15 min at 13,500 rpm to pass solutions through the centrifugal filter apparatus. These filters are expected to remove particles >90 nm per the Pall Corporation Selection Guide. Hence, the microbes and associated Cs should be retained in the filter apparatus. In this manner, the unfiltered sample represents total Cs (dissolved and microbe-associated) and the filtered sample represents only dissolved Cs. After centrifugation, an aliquot was removed and prepared for ICP-MS in 2% HNO_3 (Optima grade, Fisher).

2.5. ICP-MS methodology

All Cs measurements were conducted using an Agilent 7900 ICP-MS (Santa Clara, CA; USA) equipped with an Agilent ASX-500 series ICP-MS auto-sampler. Calibration of the ICP-MS against prepared Cs solutions from a commercial standard (1000 ppm in 2% HNO_3, High Purity Standards, Charleston, SC; USA) consistently employed six points of response measurement and resultant linearity values were frequently better than 0.9999. Indium was utilized as an internal standard throughout experiments via direct addition to each sample with the same stock solution (1000 ppm in 2% HNO_3, High

Table 1
Summary of initial (8 h injection at 0.013 mL/min) and secondary injections (10 h at 0.013 mL/min) for miniature column experiments. Note: columns conducted in duplicate.

Type	Initial Injection[a,b]	Secondary Injection[a,b]
Negative Control	5000 ppb Cs	Brine only
Stationary Phase	200 ppb Cs + stressed *or* viable microbes[c]	Brine only
Active Growth	5000 ppb Cs + log-phase microbes	Brine only
Grown with Cs	Microbes grown with Cs + spike 5000 ppb Cs	Brine only

[a] All prepared with 15% NaCl +3 mM $NaHCO_3$.
[b] The pH of mixed suspensions were measured at ~8.3 prior to injection.
[c] Concentration of microbes was approximately 10^8 cells/mL.

Purity Standards, Charleston, SC; USA) used for all samples in each run to ensure accuracy. The approximate limit of detection (LOD) across all analyses was 1.5 ppb.

3. Results and discussion

3.1. Interaction of Cs with dolomite

Results for the negative control column investigating the interaction of Cs with dolomite in the absence of *Chromohalobacter* are shown in Fig. 1. The y-axis represents the total concentration of Cs in the aqueous phase normalized with respect to the initial injection concentration as a percentage. These data suggest that Cs did not adsorb onto the bulk surfaces of the dolomite and traveled unretarded with the aqueous phase through the columns. Comparison with a ReO_4^- tracer column confirms behavior consistent with a non-reactive tracer (*Supplementary Materials* Fig. S3). Further, the similarity of unfiltered and filtered data, shows that Cs was likely not associated with a mobile colloidal fraction of dolomite in the columns. Previous studies have also found colloid concentration to be minimized in high ionic strength systems (Degueldre et al., 2000). Therefore, our observation of negligible colloid formation is consistent with previous literature. The lack of formation of mobile colloids may be due to the high activity of Na^+ ions in the high ionic strength solution which may have prevented stabilization of dolomite colloids in the effluent.

In previous experiments conducted by Chen et al., colloid-facilitated transport of Cs occurred in columns packed with natural colloids consisting mostly of clay minerals flushed with a synthetic groundwater [flushed with 1.4 or 2.8 mol/kg NaOH, 0.125 or 0.25 mol/kg $NaAlO_4$, and 3.7 mol/kg $NaNO_3$] (Chen et al., 2005). In addition, Brady et al. previously reported some sorption of Na^+ to dolomite as it interfered with Ca^{2+} and Mg^{2+} adsorption (Brady et al., 1999). The data in this research suggest that adsorption of Cs did not occur for dolomite in the bulk or colloidal size fraction. This may have occurred due to the following conditions (1) low ion exchange capacity of dolomite, (2) high ionic strength of the background solutions (15% NaCl by weight), and (3) relatively fast flow rates leading to short interaction periods.

3.2. Transport of Chromohalobacter sp. strain PZ13 in the presence of dolomite

Recovery of *Chromohalobacter* in dolomite columns was generally greater than 80% for microbes grown in the stationary phase (Figs. 2 and 3). However, actively growing microbes had a slightly decreased recovery near 60% (Fig. 4). We suggest that the actively growing microbes may have been slightly larger and, therefore, were removed due to physical straining. The higher recovery rate of microbes grown in the stationary phase may also have been influenced by cell surface properties. Previous researchers have noted that cell size and surface properties are related to growth activity with different adsorption and transport behavior due to changes in size, surface charge, and hydrophobicity (Gargiulo et al., 2007; Zhong et al., 2017). The surface charge of bacterial cells is usually negative which can imply that a Ca^{2+} bridged bond between the dolomite and the bacteria could have formed (Zhong et al., 2017; Kim et al., 2009). This may have resulted in retention of the bacterium in the porous media (Zhong et al., 2017; Gargiulo et al., 2007).

Walker et al. noted greater deposition of stationary phase *Escherichia coli* as compared to actively growing in contrast to our results (Walker et al., 2005a; Walker et al., 2005b). These results suggested that the surfaces *of E. coli* cells in the active-growth state had both a higher charge density and uniform distribution of charged functional groups than in the stationary phase. Therefore, the higher charge distribution would cause electrostatic repulsion which would yield less adhesion (Zhong et al., 2017). However, previous research by Gargiulo and team reported that *Deniococcus radiodurans* transported differently through sand-packed columns in low ionic strength systems, depending on the phase of cell growth, potentially due to greater hydrophobicity in the exponential growth phase as compared to the stationary phase (Gargiulo et al., 2007). Previous researchers suggested that the primary removal mechanism of bacteria in unsaturated columns was straining (80%) for actively growing bacteria which coincides with results in this research (Gargiulo et al., 2007). Although the experiments in this research were conducted under higher ionic strength conditions for the halophilic bacterium as compared to

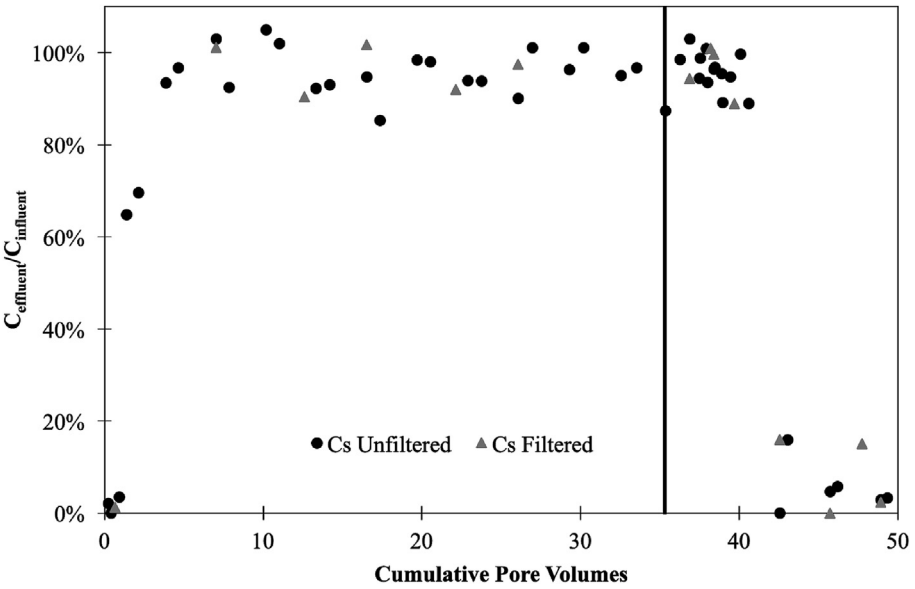


Fig. 1. Cesium effluent recovery with respect to cumulative pore volumes (one pore volume ~ 0.45 mL) for initial injection of 5000 ppb of Cs in 15% NaCl with 3 mM $NaHCO_3$ at pH 8.3 into dolomite miniature columns with a flow rate of 0.013 mL/min with unfiltered recoveries (*circles*) and filtered (*triangles*). Note: The solid line delineates the initial (*brine + Cs*) and secondary (*brine alone*) injections.

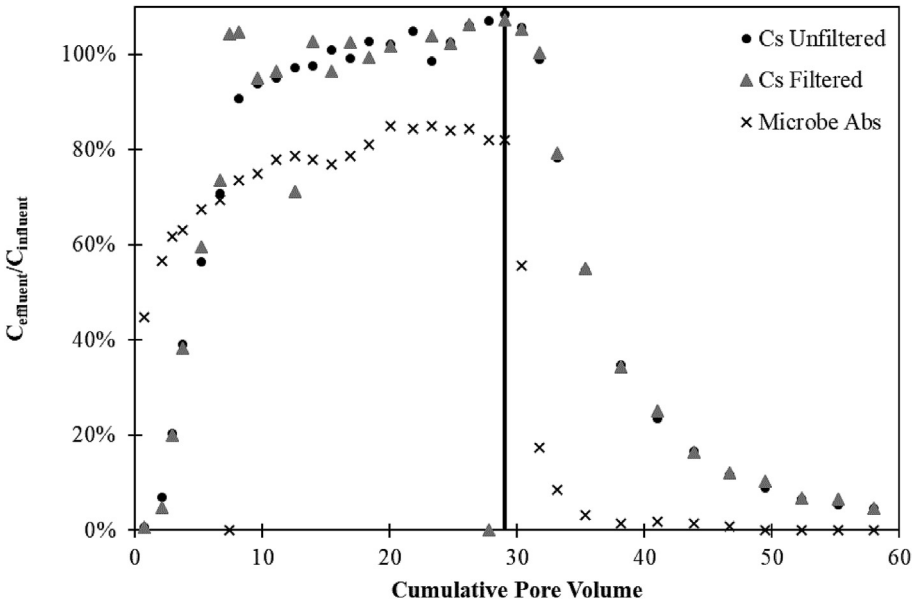


Fig. 2. Cesium effluent recovery with respect to cumulative pore volumes (one pore volume ~ 0.43 mL) for initial injection of 200 ppb of Cs in 15% NaCl with 3 mM $NaHCO_3$ with stationary phase, viable microbes at pH 8.3 into dolomite miniature columns with a flow rate of 0.013 mL/min with unfiltered *(circles)* and filtered (*triangles*) recovery of Cs and microbe recovery (*X's*). Note: The solid line delineates the initial (*brine + Cs + Chromo*) and secondary (*brine alone*) injections and error bars are based on triplicate analysis via ICP-MS.

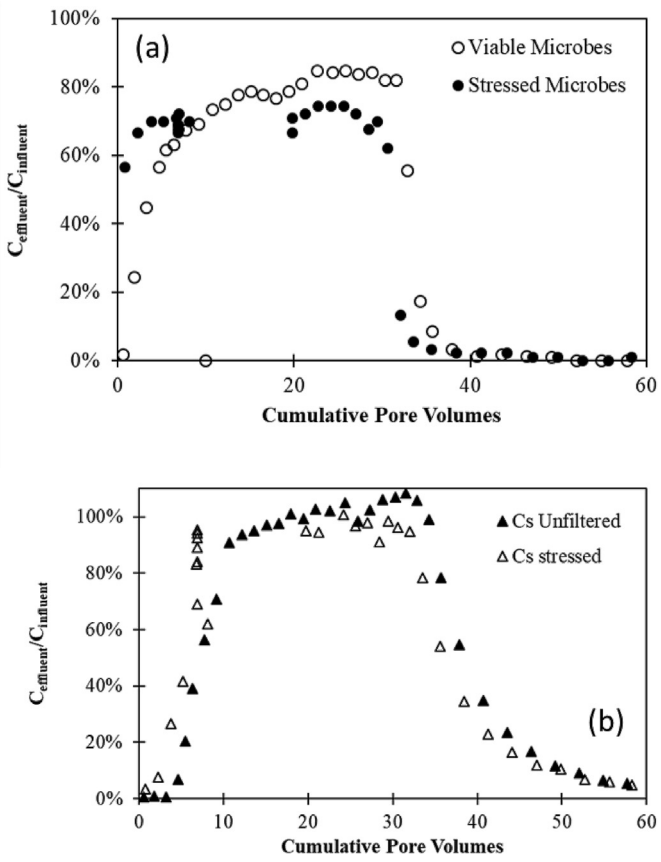


Fig. 3. Cesium effluent recovery with respect to cumulative pore volumes (one pore volume ~ 0.43 mL) for initial injection of 200 ppb of Cs in 15% NaCl with 3 mM $NaHCO_3$ with stationary phase viable *(filled)* or stressed (*unfilled)* microbes at pH 8.3 into dolomite miniature columns with a flow rate of 0.013 mL/min. The top *((a), circles)* is representative of microbial recoveries while the bottom *((b), triangles)* are the cesium recoveries with respect to the viable versus stationary experiments.

Gargiulo's research, we suggest this as the primary mechanism.

3.3. Effect of stationary-phase Chromohalobacter sp. strain PZ13 on Cs mobility

There was negligible association between Cs and the microbes in the stationary phase experiments. Fig. 2 shows that Cs breakthrough with and without filtration overlaps, implying that there was no association of Cs with the microbes as the filtration step removed *Chromohalobacter* from the aqueous phase. Therefore, any remaining Cs measured following filtration was not associated with the microbes. Further, the two series did not correlate with the optical density (OD) measurements for the microbes, which further solidified that there was no adsorption of Cs to the surface of, or intracellular uptake into, *Chromohalobacter* in the stationary phase. These results indicated that the stationary phase microbes did not have a significant effect on the mobility of Cs since (1) there was no association and (2) Cs demonstrated similar behavior to a tracer. Additional experiments were conducted to consider the impact of stressed and healthy microbes in the stationary phase with similar results highlighting a lack of interaction of Cs with the microbes (Fig. 3).

3.4. Effect of actively-growing Chromohalobacter sp. strain PZ13 on Cs mobility

Additional experiments were next conducted with actively growing *Chromohalobacter* because it was hypothesized that the intracellular uptake of Cs may not occur in the stationary phase as the microbes were not actively metabolizing. Active transport proteins for K (and thus Cs) may be differentially expressed between actively growing cultures compared to stationary. The log phase growth results imply that there was no interaction of Cs with the microbes after a 1-h equilibration of microbes and Cs prior to injection into the columns (Fig. 4). In Fig. 4, both breakthrough curves for the Cs unfiltered and filtered recoveries do not align with the microbial absorbances which, combined with strong

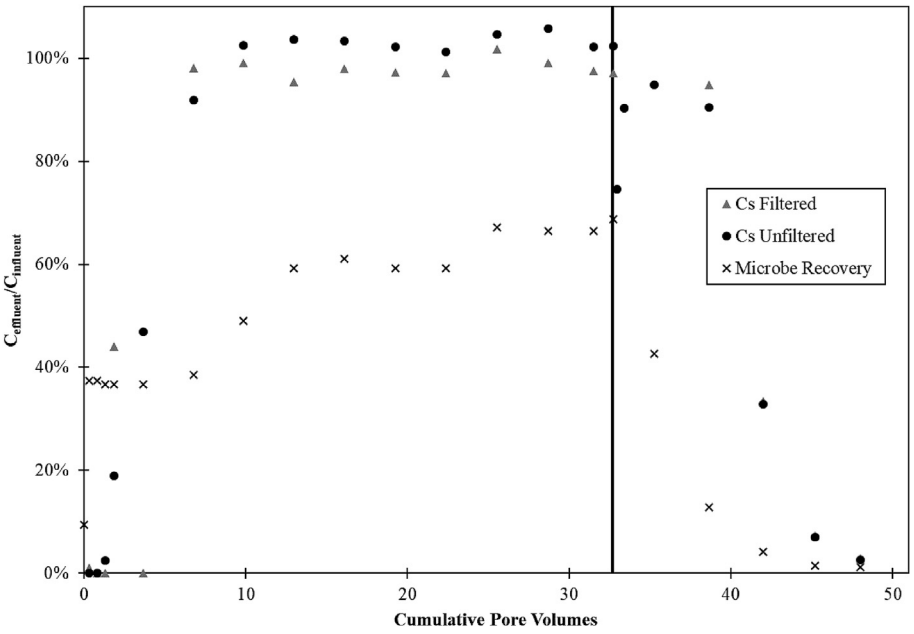


Fig. 4. Cs effluent recovery with respect to cumulative pore volumes (one pore volume ~ 0.43 mL) for initial injection of 5000 ppb of Cs in 15% NaCl with 3 mM $NaHCO_3$ with actively-growing microbes at pH 8.30 into dolomite miniature columns with a flow rate of 0.013 mL/min with unfiltered recoveries (*circles*), filtered (*triangles*) microbe absorbances (*X series*). Note: The solid line delineates the initial (*brine + Cs + Chromo*) and secondary (*brine alone*) injections and error bars are based on triplicate analysis via ICP-MS.

correlation of the filtered and unfiltered Cs results, suggests that there was neither adsorption nor uptake of Cs by the microbes. These results follow a similar trend to those with microbes in the stationary phase (Fig. 3). Further, similar results were observed with microbe cultures grown in the presence of Cs and then spiked with additional Cs with a subsequent 1 h re-equilibration (*Supplementary Materials,* Fig. S4) and in growth batch experiments (*Supplementary Materials*, Fig. S5). Both experiments yielded near equivalent results where no Cs interaction with the microbes occurred.

Previous studies have reported uptake of Cs by microbes through a similar pathway as K^+ (Avery, 1996; Bossemeyer et al., 1989; Johnson et al., 1991; Tomioka et al., 1992, 1994). In studies by Tomioka et al., an autoradiographic technique identified that two *Rhodococcus* spp. were able to accumulate Cs^+ intracellularly, with the *Rhodococcus erythropolis* strain taking up 90% of the added Cs in 24 h to reach 92 mg Cs/g of cells. These results indicated that K^+ and Rb^+ inhibited Cs accumulation which suggested that Cs can be taken up through the potassium transport system (Francis, 2012; Tomioka et al., 1994). However, previous research also reported that ^{137}Cs recoveries in microbes were much lower in samples from the river (which were rich in potassium) than in samples with deionized water (Tomioka et al., 1994). Further, whenever additional potassium was added into samples that contained deionized water, less ^{137}Cs was taken up.

Therefore, it is likely that the K concentration with respect to Cs was critical for the uptake of ^{137}Cs. Although microbes were washed prior to re-suspension in NaCl solutions for column experiments, it is possible that ample intracellular K^+ was still present to impede Cs^+ uptake. However, this does not explain the enhanced growth that was observed in the presence of Cs^+ as high as 100 mM (*Supplementary Materials,* Fig. S6). This research utilized relatively low concentrations of Cs in comparison to Bossemeyer's work. However, it is possible that high Cs concentrations are required to compete with K or other cations and lead to enhanced uptake of Cs. Bossemeyer et al. also observed slow Cs uptake with greater ratios of K in *Escherichia coli* (Bossemeyer et al., 1989). *Chromohalobacter* was grown in approximately 27 mM K^+ in these experiments.

These data suggest a less specific uptake mechanism that may be inhibited by other similar cations. The high concentration of competing cations, such as Na^+ which are required for the survival of this halophilic microbe, may also compete for potential sorption sites and membrane transport systems. Low Cs uptake in the freshwater microalga *Chlorella emersonni* was reported previously due to elevated NaCl concentrations which led to competition between Cs and Na ions (Avery, 1996). These results correlate with the results of our experiments suggesting the potential for competition between Cs and similar cations including K and Na. Therefore, the impact of total ionic strength should also be considered, especially in these experiments in 2.6 M NaCl as it may also affect the potential for Cs uptake. Further, the most closely related microbe present in genome databases, *Chromohalobacter salexigens*, possesses Trk proteins for K transport which are a low to medium affinity, meaning that they are less specific for K and can take up other ions as well. Therefore, it is logical that a microbe that thrives in hypersaline environments may not need a specific, high affinity transporter mechanism for K and may inadvertently take up similar ions including Na.

4. Conclusions

Both miniature column and growth batch experiments were effectively used to probe interactions of Cs with dolomite and *Chromohalobacter*. The high Cs recoveries in the filtered, aqueous phase is indicative of negligible interaction between the dolomite and microbes in this system. Further, the results of these experiments show that Cs was not taken up by or adsorbed onto *Chromohalobacter* despite different physiological stages and variable conditions. In the actively growing phase, the microbial recoveries were much lower than the stationary phase. The differences between the active-growth and stationary phase microbial recoveries are likely due to differences in size and surface charge properties. These bacterial transport experiments improve our understanding of contaminant interactions in high ionic strength systems.

5. Environmental implications

Given that waste accumulation and cleanup will continue to be a concern for radioactive materials created during production of nuclear power and former development of nuclear weapons, risk assessments must be consistently updated with new research. Bacterial transport in high ionic strength systems is relevant for a wide range of systems including salt repositories for radioactive waste disposal and our oceans. These results show that *Chromohalobacter* sp. strain PZ13 can be mobile in high ionic strength systems in the presence of dolomite. Although we did not observe strong adsorption or uptake of Cs into these microbes, there may be potential for uptake in systems depleted in K although it is likely that other monovalent cations can impact uptake as well. Further, the potential for transport of microbes as biocolloids has been shown to be important for other radionuclides of concern including U, Pu, and Np (Ams et al., 2013; Francis, 2012), although it was not relevant for Cs in this system.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.chemosphere.2019.06.222.

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