



# Microbially Induced Calcite Precipitation based Sequestration of Strontium

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## Introduction.

Contamination by radioactive strontium (<sup>90</sup>Sr) is a significant environmental problem. Calcite forming bacteria has also been the basis for a promising *in-situ* containment method for sequestration of divalent radionuclide and trace metal ions. In this study, ten strains were selected and identified by 16S rDNA sequencing. One of these latter stains, *Sporosarcina pasteurii* WJ-2 was further selected for subsequent study. Microbially induced calcite precipitation (MICP) by *S. pasteurii* WJ-2 strain was evaluated for its potential to counteract Sr contamination in column experiments using natural sand. *S. pasteurii* WJ-2 induced Sr containment by successfully sequestrating approximately 80% of Sr from the soluble fraction of sand.

## Methods.

1. **Isolation and identification**: Urease producing bacteria were isolated from the abandoned expressway sites (Gangwondo, Korea). Then 16s rDNA sequencing was analyzed.

2. **Urease activity assay**: The determination of the urease activity was based on the phenol-hypochlorite assay method.

3. **Sr sequestration of sand:** Sand slurry containing killed or live cells was packed into a 50 mL plastic syringe column. Columns were fed once by gravity with 20 mL of the urea and calcium chloride solution. The columns were set aside for 24 h to allow maximum crystal growth and to let excess water drip. The columns were then dried at 45°C for 24 h to facilitate evaporation of excess water. Columns were added by gravity with 20 mL of SrCl<sub>2</sub>. The Sr concentrations of the eluents were analyzed by inductively coupled plasma mass spectroscopy (ELAN 6100, PerkinElmer, USA).

### Results.

### 1. Isolation and identification

Table 1. Identification of several strains by 16S rRNA sequencing			
NO.	Area	strains	Homologous microorganism
1	WonJu, GangWon	WJ-1	Sporosarcina luteola
2	WonJu, GangWon	WJ-2	Sporosarcina pasteurii
3	WonJu, GangWon	WJ-3	Sporosarcina pasteurii
4	WonJu, GangWon	WJ-4	Sporosarcina pasteurii
5	WonJu, GangWon	WJ-5	Sporosarcina pasteurii
6	WonJu, GangWon	WJ-6	Lysinibacillus sp.
7	WonJu, GangWon	WJ-7	Lysinibacillus sp.
8	WonJu, GangWon	WJ-8	Lysinibacillus sphaericus
9	WonJu, GangWon	WJ-9	Lysinibacillus sphaericus
10	WonJu, GangWon	WJ-10	Lysinibacillus sphaericus

## 2. Urease activity assay

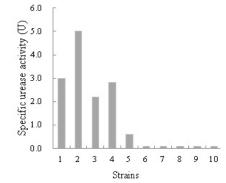


Fig.1 Specific urease activity of isolated strains. One unit of urease is defined as the amount of enzyme hydrolyzing 1 µmol urea min-1.

### 3. Sr sequestration of sand

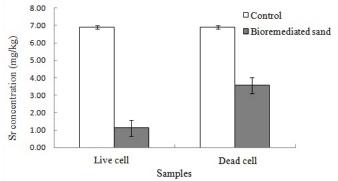


Fig.2 Sr concentration in the residual solution for dead bacterial (WJ-2) treated sand compared live bacterial (WJ-2) treated sand.

**Conclusions.** The bacterial isolate produced a significant amount of calcite precipitating enzyme, urease. Further effectiveness of MICP was demonstrated for successful sequestration of Sr.

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