

# Supplementary Material

## Evaluation of *Haloferax mediterranei* Strain R4 Capabilities for Cadmium Removal from Brines

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**Table S1.** Microorganisms with bioremediation potential for different metals. Note: NS=Non stated.

Specie	Metal	Concentration	Field	Description	Reference
<i>Chlamydomonas</i> sp.	Cd (II)	< 0.8 mM	Organisms isolated from Cd-contaminated sites; <i>Ex situ</i> laboratory assay	Tolerance and accumulation of Cd; The higher Cd concentration and the larger exposure time result in a higher accumulation of Cd	[1]
<i>Thiobacillus ferrooxidans</i> DSM 583	Cd (II)	1 M	<i>Ex situ</i> laboratory under way of being implemented in the treatment of real industrial discharges	Growth decreases when [Cd] ≥ 50mM; It accumulates Cd at acidic pH (1.6-6)	[2]
<i>Halococcus salifodinae</i> BK6	Cd (II)	2 mM	<i>Ex situ</i> laboratory assay	Gradual reduction of growth and pigmentation when Cd concentration increases	[3]
<i>Haloferax volcanii</i> BBK2		1 mM		Growth reduces when [Cd] ≥ 0.5mM	
<i>Haloarcula japonica</i> BS2		2 mM		Gradual reduction of growth and pigmentation when Cd concentration increases	
<i>Halorubrum</i> sp. BS17		< 0.5 mM		There is not tolerance to Cd at tested concentrations	
<i>Haloferax volcanii</i> BBK2	Cd (II)	1 mM	<i>Ex situ</i> laboratory assay	Tolerance to 4mM of Cd showing a significant growth reduction; The maximum accumulation of Cd was at [Cd] = 0.5mM	[4]
<i>Pseudomonas stutzeri</i> LA3	Cu (II)	400 mg L <sup>-1</sup>	<i>Ex situ</i> laboratory assay	Removal percentage is indirectly proportional to Cu (II) concentration in the medium; Highest removal percentage was at [Cu (II)] = 50 mg L <sup>-1</sup>	[5]
<i>Thermococcus gammatolerans</i> Ej3	Cd (II)	2 mM	<i>Ex situ</i> laboratory assay	Cd tolerance is caused by the coding of about a hundred genes that are expressed 120 minutes after Cd exposure	[6]
	Co (II)	2 mM			
	Zn (II)	2 mM			
	Ni (II)	0.5 mM			
	Cu (II)	0.5 mM			
	As (II)	0.25 mM			
<i>Methanosarcina acetivorans</i> C2A	Cd (II)	100 µM	<i>Ex situ</i> laboratory assay	Cd induces methanogenesis	[7]

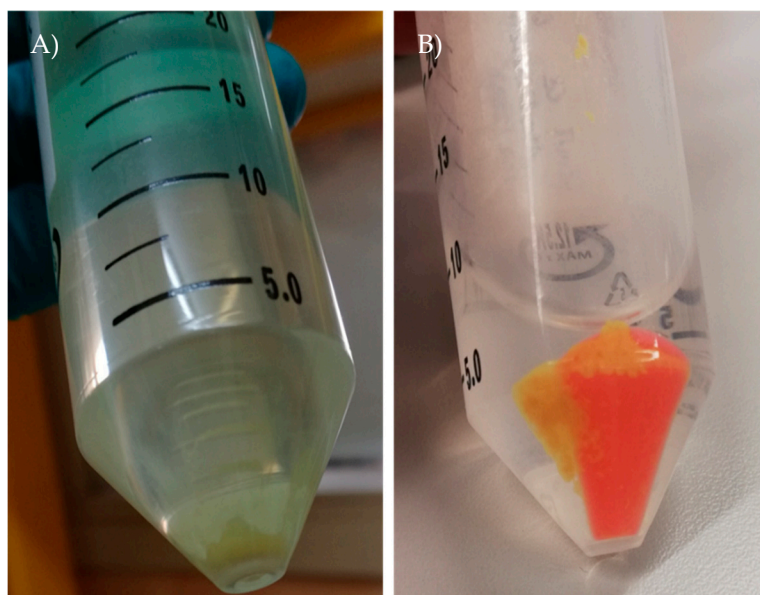
<i>Sulfolobus metallicus</i> DSM 6482	Cd (II)	5 mM	<i>Ex situ</i> laboratory assay	ATPases efflux heavy metals outwards the cell to detoxify the intracellular environment	[8]
<i>Sulfolobus solfataricus</i>	Cu (II)	>200 mM			
	Cd (II)	< 0.05 mM			
	Cu (II)	NS			

**Table S2.** ICP-OES and ICP-MS operating conditions.

	ICP-OES	ICP-MS
Plasma forward power (W)	1400	1550
Argon flow rate (L min <sup>-1</sup> )		
Plasma	15	15
Auxiliary	1.50	0.90
Nebulizer	0.70	1.09
Sample uptake rate (mL min <sup>-1</sup> )	0.5	0.3
View mode	Axial	-
Cell gas		He
Cell gas flow (mL min <sup>-1</sup> )		4
Number of replicates	3	3
Scanning mode	-	Peak Jump
Points per Peak	-	3
Dwell time (μs)	-	150
Number of sweeps	-	40
Element (wavelength, nm) / Nuclide	Ca (422.673); Cd (226.502); Fe (238.204); K (404.721); Mg (285.213); Mn (257.610); Na (588.995); Zn (213.857)	<sup>27</sup> Al <sup>±</sup> ; <sup>59</sup> Co <sup>±</sup> ; <sup>63</sup> Cu <sup>2±</sup> ; <sup>60</sup> Ni <sup>±</sup> ; <sup>88</sup> Sr <sup>±</sup> ; <sup>137</sup> Ba <sup>±</sup>
Internal standard	Sc (391.182, 361.383)	<sup>101</sup> Ru <sup>±</sup>

**Table S3.** Summary of ANOVA results for elemental variation analysis according to the different Cd treatments (0, 0.2 and 0.4 mM of Cd (II)).

Source of variance	F-value (F <sub>2,6</sub> )	p-value	Tukey HSD test
Al	0.002	n.s.	-
Ba	1.529	n.s.	-
Ca	30.51	n.s.	-
Co	0.647	n.s.	-
Cu	1.146	n.s.	-
Fe	1.465	n.s.	-
K	5.802	< 0.05	0 < 0.2; 0.2 = 0.4; 0 = 0.4
Mg	6.035	< 0.05	0 = 0.2; 0.2 = 0.4; 0 > 0.4
Mn	98.730	< 0.001	0 < 0.2 < 0.4
Na	4.473	n.s.	-
Ni	1.885	n.s.	-
Sr	3.709	n.s.	-
Zn	114.300	< 0.001	0 > 0.2 > 0.4



**Figure S1.** Appearance of the yellow precipitate CdS in CM. **A)** No centrifuged CM without cells and **B)** centrifuged CM with cells. Note: pink pellet corresponds to the cell biomass.

## References

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