

## AN INNOVATIVE APPROACH TO BIOREMEDIATION OF MERCURY CONTAMINATED SOILS

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### Introduction

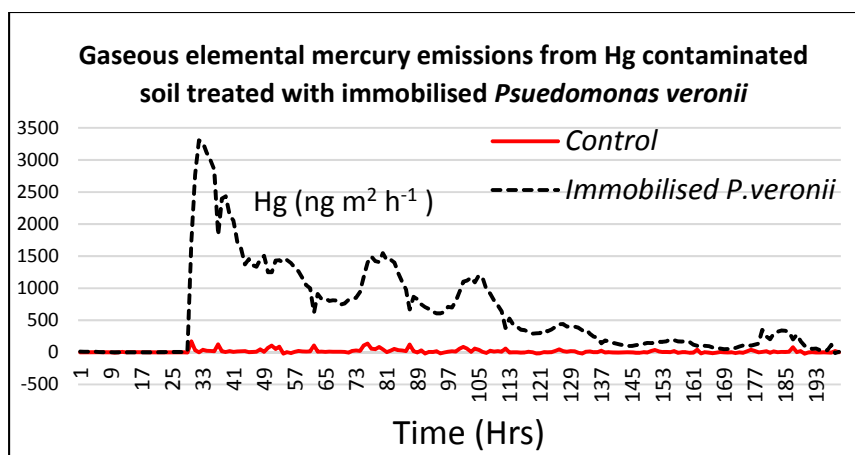
Mercury (Hg) is a highly toxic contaminant from mining and industrial operations, and is subject to complex biogeochemical cycling once released into the environment. Hg contaminated soil remediation has proven expensive and or technically challenging. Microbial tolerance of Hg is well understood but not well explored as a viable solution to soils contamination. One bacterial strategy mediated by the *mer* operon involves uptake of Hg<sup>2+</sup> ions followed by intracellular enzymatic reduction by mercuric reductase to neutral Hg<sup>0</sup>, whereupon it volatilises and passively diffuses from the cell and out of soil. Augmenting soils with beneficial bacteria is a well understood *in situ* bioremediation concept but delivery of microbial loads heterogeneous and remote sites proves technically challenging. This research utilised a biopolymer to immobilise Hg volatilising bacteria to a bulk natural substrate which was stored, transported internationally, and applied to Hg contaminated soils. The resultant Hg soil-atmospheric flux was monitored, as was ongoing shelf viability and functionality of immobilised cells. Results indicate this method has great potential in providing a storable, readily transportable and inexpensive product that could be applied to *in situ* bioremediation.

### Methods

Live *Pseudomonas veronii* cells grown to stationary phase were immobilised in a xanthan gum based biopolymer via encapsulation and subsequent mechanical coating on sterilised natural Australian zeolite granules (2-4 mm, clinoptilolite with trace modernite), as previously described (Johnson et al., 2001). The granules were dried and stored at room temperature for ten weeks. Viability was monitored by weekly CFU enumeration. One batch was transported from Australia to the United States of America where it was applied in a ratio of 1:1 v/v to soil heavily contaminated with Hg from a heap leaching operation. The control was contaminated soil inoculated with sterile zeolite. Hg flux was measured by atomic absorption using a Tekran 2573A Hg Analyser (0.1 ng m<sup>-3</sup> detection limit) in conjunction with a Tekran Automated Dual Sampling Unit (TADS) linked to cylindrical dynamic flux chambers (TADS) using PFA Teflon tubing, as previously described (Eckley et al., 2010).

### Results

The cells retained both viability and Hg volatilisation functionality. Maximum flux rates exceeded 3 μg Hg m<sup>2</sup> h<sup>-1</sup> from soils containing 8 mg kg<sup>-1</sup> Hg treated with the zeolite granules containing immobilised cells (Figure 1). This flux rate was ~3 orders of magnitude above background levels. Speciation data revealed a concomitant drop in gaseous oxidised mercury (GOM) flux. Cells displayed good storage capacity under ambient room conditions (~1.0 x 10<sup>-8</sup> CFU mL<sup>-1</sup> starting concentration, rapidly stabilising at ~1.0 x 10<sup>-6</sup> CFU mL<sup>-1</sup>).



**Figure 1.** GEM emissions after application of immobilised *P.veronii* encapsulated in biopolymer bound to zeolite at time zero + 29 hrs. Background emissions were positive flux of about  $2 \text{ ng m}^2 \text{ h}^{-1}$ . The application of immobilised cells increased flux rate by  $\sim 3$  orders of magnitude to beyond  $3000 \text{ ng m}^2 \text{ h}^{-1}$  initially.

### Conclusion

This is the first research the authors are aware of that uses immobilised cell technology for Hg volatilisation for bioremediation. The greatly increased rate of Hg emissions indicates treatment with Hg volatilising organisms offers great potential for liberation, extraction and capture of emitted gaseous elemental mercury (GEM), which can then be stored or recycled appropriately as metallic Hg. The immobilising excipients used in this research offer potential to overcome many logistical issues with delivery of suitable microbial loads to locations of mercury contamination, and represents a facile and inexpensive method of augmenting sites with microbial consortia for bioremediation.

### References

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