

## Metal utilisation in *Clostridium* microbial biocatalysts

"For GBL, this study highlighted the importance of metal requirements for commercial and research and development processes."

Durham GreenBiologics

Peter Chivers, Durham University; Liz Jenkinson, Green Biologics

**OUTCOMES:** Metal contents were measured at different times during a GBL fermentation protocol used for Research and Development. The ICP-MS analysis revealed a large increase in metal content in *Clostridium*, coincident with the onset of butanol production. To complement the experimental data, bioinformatics analyses identified ten candidate metal sensor genes. These genes encode regulator proteins responsible for sensing and responding to changes in metal content. In several cases, their regulatory targets have also been identified. The proteins encoded by these genes have been overexpressed in *E. coli* to facilitate future studies of their metal selective transcriptional responses. These results will enable more detailed analysis of metal homeostasis networks in *Clostridium* to understand their link with butanol production or competing processes.



SDS-PAGE analysis of overexpression of Clostridium metal sensor proteins in E. coli

**INITIAL AIMS:** Little is known about the metal-demands of *Clostridium* strains during solvent production. This project will explore the metal requirements of solventogenic *Clostridium* during commercial and research and development processes used by Green Biologics. Metal content will be analysed by ICP-MS. A complementary aim is to identify the metal sensor genes responsible for maintaining metal homeostasis so that they may be overexpressed in *E. coli* to define the metal selective response of each protein.

 Understanding metal demands could lead to improved control of fermentation processes







Biotechnology and Biological Sciences Research Council