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A proteomic approach to optimizing gas fermentation in industrially relevant acetogens

PI Professor Phillip Wright, University of Sheffield

There is relevant interest, from the economic and environmental point of view, on certain bacteria that are able to grow on waste gases such as syngas, including the greenhouse gas carbon dioxide and the poisonous gas carbon monoxide instead of sugars. In doing so, these bacteria can convert the gases into useful biofuels (such as ethanol) and also help to remove harmful gases from the environment. These waste gases are generated at high volume in major industrial processes such as steel and petroleum production.

Therefore, we have investigated the anaerobic bacterium *Clostridium autoethanogenum* DSM10061 as a waste gas utiliser (grows on carbon monoxide) and biofuel (e.g. ethanol) producer. Different growth conditions and feedstock gas compositions were tested to examine the impact on biofuels production and on the health/viability of the bacterium. Using mass spectrometry (an analytical technique that identifies chemicals by accurately determining their mass), we were able to identify key proteins that play major roles in growing on these gases and how they help the cells produce biofuels. Proteins acting together in so-called metabolic pathways catalyse the reactions in a cell that allow the cell to live and thrive as well as produce beneficial products. We evaluated these metabolic pathways to understand which condition is better at making a particular biofuel compared to another. We generated metabolic maps to show these differences in behaviour. We were able to show that higher levels of carbon monoxide and/or ethanol in the feed reduced the cells' capability to produce more biofuels.

This finding, coupled with the data and metabolic maps, will allow us to design and engineer/rewire better *Clostridium autoethanogenum* cells that are more resilient to high concentrations of carbon monoxide, for example, in the feed whilst simultaneously being able to generate higher amounts of biofuels such as ethanol or butanol.