CBMNet
CROSSING BIOLOGICAL MEMBRANES NETWORK, A BBSRC NIBB

The University Of Sheffield.

UNIVERSITY of York

BBSRC

Condition-specific engineering of Pseudomonas metabolism

The Challenge

Microorganisms have a high capacity of synthesizing a wide range of surface-active compounds, generally called biosurfactants.

Rhamnolipids are a common biosurfactant and due to their non-toxic nature, excellent biodegradability, high surface/ interfacial activity, high thermal/chemical stability, production from renewable resources and the ability to form microemulsions, rhamnolipids can be used as emulsifiers and stabilizers in a great number of sectors.mEnhanced production of rhamnolipids is key for industrial applications, ranging from cosmetics to bioremediation of organic and heavy metal contaminated environments.

Rhamnolipids are predominantly produced by the microbe *Pseudomonas aeruginosa*. However, *Pseudomonas putida* is a model organism with greater metabolic versatility and potential for industrial applications. Computational methods for metabolic engineering are able to model and optimise such biological models, leading to the improvement of a given biotechnological pipeline.

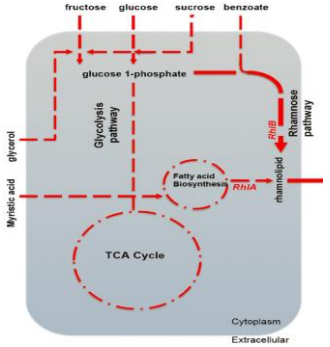
The Research

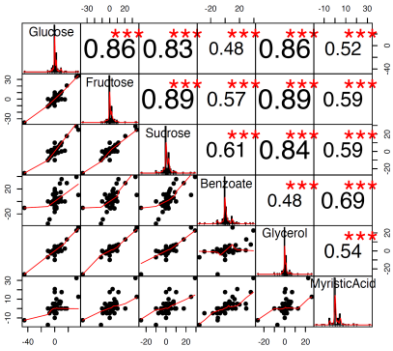
Dr Angione is a Senior Lecturer at Teesside University. The research in his laboratory focusses mainly on computational biology and modelling for biomedical applications.

TeeGene Biotech Ltd is a spinout venture from Teesside University, which is pioneering the use of biosurfactants in a range of household, environmental, cosmetic and biomedical applications.

Dr Angione applied for a CBMNet Business Interaction Voucher with TeeGene Biotech Ltd to develop a collaborative partnership and produce preliminary data that could be built on in future funded projects.

The project aimed to investigate the metabolic capabilities of *P. putida* for rhamnolipid biosynthesis using metabolic and biosynthetic engineering approaches. The analysis would be based on computational modelling and metabolic engineering.





CBMNet Business Interaction Voucher

The Result

The engineered genome-scale model of *Pseudomonas putida* built during this BIV project can already be used to predict and maximise rhamnolipid production and transport through the cell membrane.

The *P. putida* model was optimised to maximise the production and export of biomass and rhamnolipids. By introducing the non-native genes RhlA and RhlB (from *P. aeruginosa*), P. putida can produce rhamnolipids. The project found the majority of rhamnolipid production originated from the rhamnose pathway rather than from the FA pathway.

To establish the best growth condition for optimal rhamnolipid synthesis, the project investigated alternative carbon sources separately: fructose, sucrose, glycerol, benzoate and myristic acid. Findings report that metabolism of myristic acid (C-14), followed by fructose and sucrose/glucose, provided the best condition for optimal rhamnolipid synthesis.

The Future

The promising results found during this project have strengthened the collaboration between TeeGene and the Computational Biology group at Teesside University.

Results will be extended with poly-omics modelling and validated in an upcoming project for which we are seeking funding as a Proof of Concept.

The resulting pipeline will finally be used to build a genome-scale model of *Pseudomonas teessidea*, first isolated and characterised by TeeGene, for which a model is unavailable.

The resulting project will elucidate the metabolic engineering steps for overproduction of rhamnolipids and their transport out of the cell membrane.

This project was funded through the Crossing Biological Membranes Network (CBMNet) by the Biotechnology and Biological Sciences Research Council (BBSRC)

Email: cbm@sheffield.ac.uk
Telephone: 0114 222 9766
Website: www.cbmnetnibb.net
Twitter: @CBMNet_NIBB

"The funding awarded by CBMNet has kick-started a fruitful collaboration with TeeGene Biotech. This collaboration has been extremely successful and has already generated a number of additional interdisciplinary projects, blending computational and mathematical tools with industrial biotechnology expertise."

Claudio Angione
Teesside University

"This BIV work with Teesside University uncovering some of the challenges we phase over the years to identify and optimise rhamnolipid production. This work integrate TeeGene's core technologies and business aspirations with biotechnology and bioprocessing expertise at Teesside to create a central enabling technology for biomanufacturing of biosurfactants."

Pattanathu Rahman
TeeGene Biotech Limited