# CBMNet Proof-of-Concept Award:

## Viral antigen production for vaccine-mediated disease prevention by rational glyco-engineering-mediated protein secretion in a cell factory

## The Challenge

Flaviviruses are a family of viruses spread by mosquitos, which cause a range of diseases such as dengue fever and zika microencephaly. Global human movements, and expanding mosquito habitats mean that these diseases pose greater challenges than previously perceived. A vaccination approach associated with these diseases is problematic because, while affording protection to one type of a particular virus, immunization can promote more severe disease caused by one of the other viral types. The proteins that sit on the surface of all flaviviruses share some common shapes, and it is these which provoke the more severe disease form. However removing them makes immunization ineffective by disrupting the overall protein shape when it is part of a vaccine.

## The Research

Professor Carl Smythe, is a Professor of Cell Biology at the University of Sheffield. Research in his laboratory uses molecular cell biology approaches to understand quality control or surveillance mechanisms that operate in cells to ensure fidelity of function, the consequences when they fail, and how they may be exploited to ameliorate disease. Professor Smythe applied for a CBMNet Proof-of-Concept project with Excivion Limited, with the aim to implement a strategy whereby the protein that needs to be produced to create a vaccine is made in such a way, that the offending structure cannot be "observed" by the immune system, and is effectively cloaked via molecular modification. This type of modification can only be achieved by certain types of cell factories.

## The Result

The project succeeded by engineering modified Dengue and Zika viral capsid genes to produce cognate proteins, predicted to retain native protein structure, and stably introducing these genes into mammalian cell factories for subsequent isolation after secretion across the plasma membrane. This solves two production problems simultaneously - by exploiting the integral functionality of mammalian cell factories to glycosylate specific peptide motifs, generating homogeneous protein products critically containing an epitope masked by glycoengineering, while utilising the same factories to efficiently secrete the desired products for subsequent purification prior to utility as both diagnostics and vaccines. Using the biOMICS™ technology associated with the University's expertise in highly efficient protein production, together with our mass spectrometry capability enabling exquisite molecular characterisation, the project was a success.

## The Future

This work, is extremely timely. Zika is a flavivirus, closely related to Dengue virus. The only available relevant vaccine product introduced to the market (DengVaxia) which has been predicted to reach a $400M annual market, was recently withdrawn, for the very reasons anticipated in the design and development of the prototypes used in this project. The data from the project provided the collaborating company with sufficient confidence to successfully apply for a new round of Innovate UK funding (valued at £2 million) to develop a novel vaccine for the prevention of the spread of ZiKa virus and the developmental disorders it is known to cause.

**"We are delighted to have the opportunity, afforded by CBMNet, to help Excivion address this pressing problem in the development of novel biologics for therapeutic intervention. The solutions from this project, not only indicated the importance of a critical mass of interdisciplinary expertise in addressing biomedical challenges, but has provided insight into the challenges of biotech industry, providing researchers and students with insight into real-world technological problems."** Professor Carl Smythe, University of Sheffield