# Beyond AlphaFold: Experimental and computational folding of a de novo membrane protein

**Supervisory team:**
**Main supervisor:** Dr Paul Curnow (University of Bristol)
**Second supervisor:** Prof Adrian Mulholland (University of Bristol)

**Collaborators:** Prof Tomoaki Matsuura (Tokyo Institute of Technology)

**Host institution:** University of Bristol

**Project description:**
It has been suggested that the advent of AlphaFold has now "solved the protein folding problem". In fact, understanding the precise steps that a protein must take in order to adopt a specific, functional three-dimensional fold remains a very active area of research. In particular, the folding behaviour of integral membrane proteins – which constitute about one-quarter of all the proteins in nature – is still quite unclear. A deeper comprehension of membrane protein folding and assembly is very important as part of our fundamental knowledge of protein science, but also for appreciating how mis-folding might occur in human disease, and how folding processes can be exploited in protein design and synthetic biology. The Curnow lab has recently developed a man-made, synthetic membrane protein that offers a new opportunity to study folding processes at the molecular level. This unique protein can be made by living bacterial cells. It is trafficked to the bacterial cell membrane where it binds tightly to the catalytic cofactor heme. This means that we now have, for the first time, a simple and extremely robust mimic of the heme-binding bioenergetic proteins that power biology via respiration and photosynthesis.

In this PhD project, the student will combine computational and laboratory methods to understand the biosynthesis of our artificial cytochrome. The laboratory work will move out of the complex cell environment and instead use well-controlled in vitro systems to manufacture the protein. This approach allows us to break down the folding and assembly process into specific, discrete steps and so to unpick exactly what is happening at each step. It will provide an unprecedented insight into the formation of the protein-heme complex, and once the methods are established we can then apply this same technique to natural proteins too. Surprisingly, despite their critical importance to cellular life, bioenergetic proteins have not previously been closely studied in this way. The other aspect of the studentship is to build a computer model of the entire folding process. Such molecular dynamics methods have not previously been possible because of the computational demands of simulating a realistic lipid membrane. However, new methodological developments and increasing computational power now put this long-held goal within reach. This project thus offers an exciting combination of theory, experiment and simulation that will be used to address one of the remaining 'grand challenges' in protein science.

**Our aim as the SWBio DTP is to support students from a range of backgrounds and circumstances. Where needed, we will work with you to take into consideration reasonable project adaptations (for example to support caring responsibilities, disabilities, other significant personal circumstances) as well as flexible working and part-time study requests, to enable greater access to a PhD. All our supervisors support us with this aim, so please feel comfortable in discussing further with the listed PhD project supervisor to see what is feasible.**