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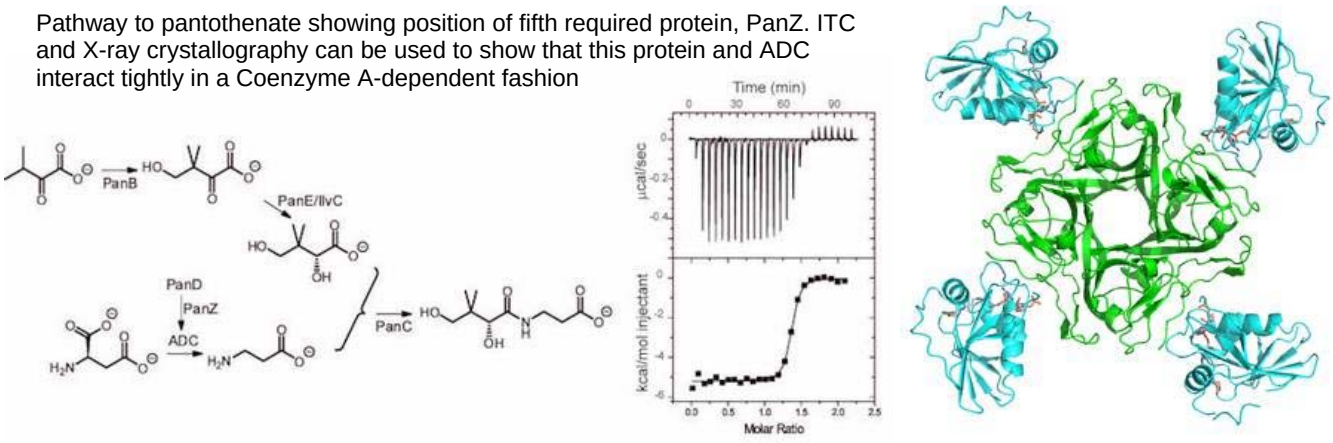
A related project to engineer the PanD/PanZ complex as a CoA sensor is available through the BBSRC DTP: https://www.findaphd.com/search/ProjectDetails.aspx?PJID=57735&LID=735

Protein-protein interactions for metabolic regulation

Protein-protein interactions are critical to the function of many signaling pathways. Their importance in the regulation of central metabolism is less well studied, however PPIs can serve to modulate the catalytic activity of enzymes. In this project, you will build on our recent discovery of a protein-protein interaction which regulates the bacterial pantothenate biosynthetic pathway. This interaction is between the enzyme aspartate decarboxylase (ADC) and its essential activating factor PanZ. This second protein is required to form the covalently-bound cofactor found in ADC. Once activated, ADC generates beta-alanine which is then used to form the coenzyme A (CoA), which is essential for central carbon metabolism and fatty acid biosynthesis. We were therefore surprised when our structural characterisation showed that CoA was essential for formation of the activation complex; it appeared to create a positive feedback loop! Subsequent studies have shown that, in fact, the protein interaction has two functions: it is required for activation, but it also inhibits the catalytic activity of ADC once it has been activated – a negative feedback loop.

Pathway to pantothenate showing position of fifth required protein, PanZ. ITC and X-ray crystallography can be used to show that this protein and ADC interact tightly in a Coenzyme A-dependent fashion



Different projects are available in this area, for example in: (i) using our knowledge of this complex to generate new potential antibiotic compounds; (ii) carrying out a more detailed (and essential) kinetic characterisation of this regulatory system and investigating its behaviour *in vivo*; (iii) determining whether this is the only example of this class of regulation in bacteria – or if it is the exemplar of a new kind of regulatory element for metabolism.

Depending upon the precise area of the project, research work will require a broad range of skills to be used including: *organic synthesis* – to synthesise candidate small molecules that could rehearse the activity of CoA in the system; *assay development* – using protein modification approaches to develop new probes of protein-protein interaction for screening; *molecular biology* and *bacterial strain engineering* – to investigate the in vivo consequences of regulation; and *protein overexpression* and purification – as a precursor to assay development, *biophysical and structural characterisation* by ITC, NMR, X-ray crystallography and SAXS. The work may be carried out in collaboration with a number of national and international collaborators including Professor Arwen Pearson (Hamburg), Professor Niki Hironori (NIG, Japan) and Dr Richard Foster (Leeds, MedChem).

References

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