

Case Study: The use of plasmid and protein purification systems in a high-throughput format for recombinant protein production

The Protein Production Facility at the University of Leeds uses Promega's Wizard® MagneSil® Plasmid Purification System and MagneHis™ Protein Purification System as part of their high-throughput workflow

Recombinant Protein Production

Recombinant production of proteins is a very powerful and commonly-used tool in research. Proteins that are not naturally produced in a certain form or in substantial quantities can be expressed and purified on a larger scale from a specific genetic template, often using living cells, for downstream studies. Recombinant proteins can be used in a wide range of applications including structural and functional studies, biotherapeutic drug discovery and screening.

The University of Leeds Protein Production Facility

Opened in 2017, the Protein Production Facility at the University of Leeds is a core facility dedicated to producing high quality recombinant material for functional and structural studies. They offer equipment, services, training and expert assistance with all aspects of protein purification for users within the university, as well as externally to academic and industrial institutions.



Dr Brian Jackson (Facility manager) at the Protein Production Facility, University of Leeds

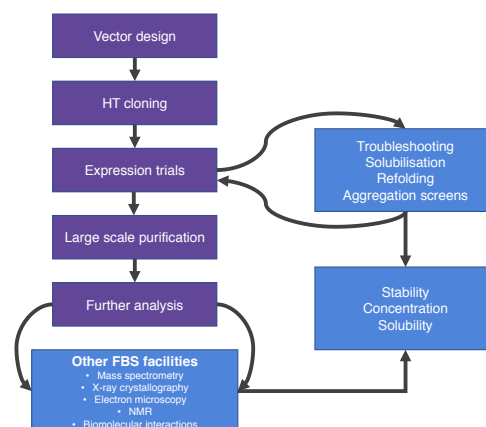
The Facility, situated in the Astbury building, was purpose-built with funds from the University of Leeds and the Royal Society Wolfson Foundation Laboratory Refurbishment Scheme to house brand new, state-of-the-art molecular biology equipment.

Workflows

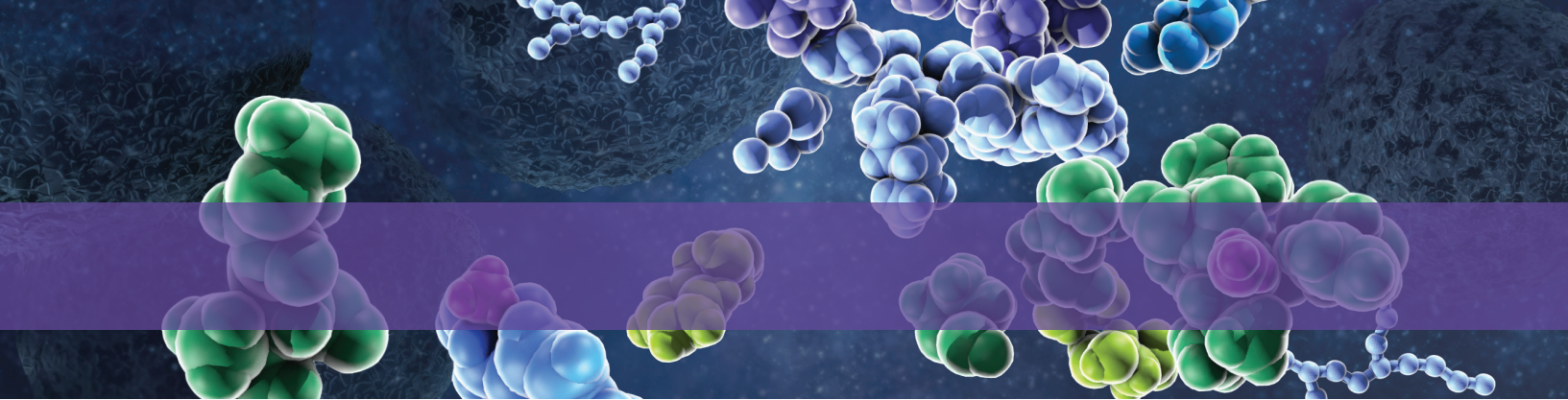
Facility manager Dr Brian Jackson said, *"The Protein Production Facility caters for all stages of the recombinant protein expression workflow, from vector design to cloning, and from small-scale expression screening to large-scale growth and purification. The workflow includes any liquid chromatography method for purification, and proteins can be expressed in bacterial, insect and mammalian systems."*

"The Protein Production Facility caters for all stages of the recombinant protein expression workflow."

Depending on the protein of interest and expression system used, the whole workflow can be performed in less than 1 month. The Facility can perform the work as a service, or researchers can be trained to use the equipment to express and purify proteins.



Protein Production Facility workflow. Stages in purple are performed directly by the Facility. Stages in blue are performed either in collaboration with, or exclusively by, other Research Facilities in the Faculty of Biological Sciences.



High-throughput chemistry for plasmid extraction and protein purification

Brian added, "An important aspect of the Protein Production Facility workflow is high-throughput cloning and protein expression screening. The Facility has a Hamilton Microlab STAR liquid handling platform for automating a number of key techniques. For example, plasmid minipreps are performed using Promega's Wizard MagneSil Plasmid Purification System, and protein expression screening is performed using Promega's MagneHis Protein Purification System, and both techniques can be automated in a 96-well format.

"For MagneSil we generally get around 20-50ng/ μ l of DNA, which is the perfect amount for checking cloning and sequencing. MagneHis yields are much more variable because we use it to screen for the best purification conditions, but when we get successful protein expression it's common for us to get in the region of 10-20 μ g of protein, although we have had much more in some cases (~100 μ g) which is more than enough for us to analyse for our downstream applications.

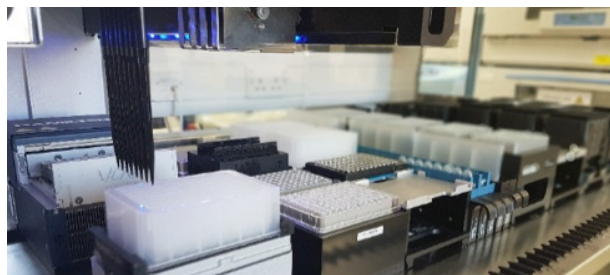


The Protein Production Facility Laboratory

"Automated method writing was handled by Promega's Field Support Scientist with unlimited assistance to ensure the workflows performed optimally. These high-throughput methods are critical for ensuring that the Facility can meet the needs of its clients."

Promega Field Support Scientists (FSS)

The FSS are a dedicated team of liquid-handling specialists who work with and provide continuing support to customers in the implementation of high-throughput extraction and quantitation chemistries on complex liquid handling systems. Promega continues to support the Protein Production Facility from both a technical and customer service aspect.



The Hamilton Microlab STAR workstation at the Facility

Promega offers several magnetic-bead based chemistries for the rapid isolation of plasmid DNA and also the purification of polyhistidine- or HQ-tagged expressed proteins in a 96-well plate format on liquid handling platforms.

Promega partners with all of the major liquid handling suppliers in the UK, and provides and supports a wide variety of enabling chemistries for plasmid purification, nucleic acid extraction, nucleic acid quantitation and assays for cell biology analysis. In addition, Promega supplies ancillary reagents and instrumentation for stand-alone benchtop nucleic acid extraction, amplification and quantification.

To learn more about Promega products, please visit: www.promega.com, or contact Promega to discuss your high-throughput project: ukcustserve@promega.com.

To find out more about the Protein Production Facility please use the University of Leeds online enquiry form: <http://www.fbs.leeds.ac.uk/facilities/protein/contact.php>