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From Gene to Protein Automatically. A Platform Technology for Systematic, Automated and High Throughput Protein Expression And Purification. Grant Cameron, Robert Mount, Kevin Auton, *NextGen Sciences Ltd.* E-mail: lorna.watson@nextgensciences.com

Keywords: Protein crystallography; Protein expression; Protein

Drug discovery and disease scientists increasingly require protein samples fit for purpose in research involving crystallography. This has led to a growing need for new tools to overcome the problems associated with protein expression. This presentation will discuss the design and development of a unique combination of biology, hardware and software tools that enable the entire process of expression vector construction, protein expression and subsequent purification to be automated. The result is a technology enabling systematic and parallel production of many hundreds of purified proteins with minimum hands-on time. A range of unique expression vector systems, which incorporate a series of fusion partners together with affinity tags for subsequent purification to enable optimal soluble protein expression, will also be discussed. The technology integrates a unique information management system for sequence, sample and protein tracking. It is possible to start with PCR products and cDNA clones and plan, schedule, automate and track the whole process of sub-cloning, expression and purification of hundreds of proteins in parallel. The use of the system will be demonstrated with the automated production of human p53 protein, and the optimisation of expression by the identification of ideal fusion constructs.

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Refolding, Purification and Crystallisation of a Malaria Antigen, AMA1 from Plasmodium falciparum. Adrian Batchelor, Tao Bai and Aditi Gupta, *University of Maryland at Baltimore, USA.* E-mail: abatchel@rx.umaryland.edu

Keywords: Refolding; Expression; Crystallisation

Malaria antigens are notoriously difficult to produce for crystallization studies for several reasons. Firstly, there is an extreme AT rich codon bias that results in cloning and expression difficulties. Secondly, malaria antigens are not N- or O- glycosylated and expression using eukaryotic expression systems is problematic because aberrant glycosylation occurs. Thirdly, malaria antigens frequently contain multiple disulphide bonds such that intracellular expression of correctly folded proteins is unlikely.

Our approach to producing malaria antigens for crystallization studies is to express insoluble protein in bacteria and to refold and purify refolded protein. We will describe the trials and tribulations of our experiences when attempting crystallize one malaria antigen, AMA1 from *Plasmodium falciparum*. This approach has ultimately been successful and we have generated high quality crystals that diffract x-rays to 1.9 Angstroms.