How new strategies can improve productivity - rMMS microseeding for crystallization and DLS for cryoEM

Patrick Shaw Stewart, Douglas Instruments Ltd, Hungerford, UK (carolyn@douglas.co.uk)

Abstract

Random Microseed Matrix-Screening (rMMS), where seed crystals are added automatically to random crystallization screens, is a significant recent breakthrough in protein crystallization [1]. During the ten years since the method was published, understanding of the theoretical advantages of the method has increased [2 - 4], and several practical variations on the basic method have emerged. Important variations that will be discussed include combining seeds from several hits [5], the best methods of selecting hits to optimize [2], and cross-seeding targets with crystals of homologous proteins [6].

Single-particle cryoEM clearly has great potential to determine the structures of macromolecules when crystallographic approaches are not available. The throughput of cryoEM is, however, low, with only one or a few samples being analysed per day. This creates a need for a pre-screening approach to investigate the behaviour of macromolecules in solution. Douglas Instruments has worked closely with Xtal Concepts to develop a screening approach were e.g. 96 wells can be analysed by DLS using only a few microlitres of sample [8, 9]. This concept will be outlined briefly, with examples.

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