

W0307

Crystallization Strategy in the Midwest Center for Structural Genomics. Youngchang Kim, Irina Dementieva, Ruiying Wu, Min Zhou, Grazyna Joachimiak, Lour Lezondra, Pearl Quartey, Hui Li, Andrzej Joachimiak, Biosciences Div., Structural Biology Center & Midwest Center for Structural Genomics, Argonne National Laboratory, 9700 S.Cass Ave., Argonne IL 60439, USA.

One of the critical rate-limiting steps in structural biology occurs between producing purified proteins and obtaining diffraction quality crystals for structure determination. Using the large amount of data obtained during implementation of the Midwest Center for Structural Genomics (MCSG) structure determination pipeline, we made an attempt to optimize these steps. The Argonne's MCSG purification pipeline was set up to produce "structural genomics grade" proteins which need to be highly homogeneous, in milligram quantities, and produced in high numbers quickly and reproducibly. Proteins, 4 – 16 different proteins at a time, are purified in a few chromatographic steps following the well-established standard protocols on a semi-automated AKTA-3D system. Taking advantage of nanoliter crystallization system, the lower limit of protein required for the complete screen consisting of number of different commercial crystallization matrices incubating at two different temperatures is less than 10 mgs and the screening can be completed in less than two hours. Majority of proteins are prepared as selenomethionine proteins with excellent yields. In the past several months nearly 600 soluble proteins purified from *Bacillus subtilis*, *B. stearothermophilus* and *Aquiflex aeolicus*. Macroscopic crystals were obtained for 35% of protein samples and more than a half of these crystals were of diffraction quality and did not require significant optimization.

This work was supported by the by grants from the National Institutes of Health (GM62414) and the U.S. Department of Energy, Office of Biological and Environmental Research, under Contract W-31-109-ENG-38