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Auto-Rickshaw: An online tool for validation of an X-ray diffraction experiment

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We present automated crystal structure determination platform, Auto-Rickshaw[1], that combines invoking of a number of macromolecular crystallographic software packages with several decision-making steps in between. A large number of structure solution paths are encoded in the system and the optimal path is selected by the decision-makers as the structure solution evolves. The Auto-Rickshaw phasing protocols include SAD, SIRAS, MAD, MR and MRSAD.The primary aim of the pipeline is to validate the crystallographic experiment at the synchrotron site while the crystal is still at or near the beamline. Thus, the system has been optimized for speed, so that typically within a few minutes the answer is provided whether the collected data will be of sufficient quality to allow successful structure determination. The EMBL-Hamburg beamline user have been successfully using this system for a little over three years. It is now available as a web serivce (http://www.embl-hamburg.de/Auto-Rickshaw) in the structural biology community. An overview of the Auto-Rickshaw pipeline with its design, functionality, decision making and some examples and the way this platform is used as a feedback system for X-ray data collection or validation of X-ray experiment, will be discussed.

Keywords: auto-Rickshaw, phasing, data validation

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AutoPROC - A framework for automated data processing

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autoPROC is a set of tools and programs to automate the steps performed during data processing: (a) determination of correct direct beam coordinates, (b) indexing of diffraction images, (c) determination of accurate cell parameters, (d) integration of a series of images, (e) scaling and merging of each sweep separately as well as of a collection of related sweeps, (f) analysis of anomalous signal, (g) automatic determination of most-likely space group symmetry. It is designed to be independent of the actual program(s) used for the various steps. The current version offers complete processing streams based on either MOSFLM [1] or XDS [2], with crossing points between them at various levels. The scaling and merging step - which is also available as a stand-alone module - has been implemented using SCALA [3]. autoPROC has been developed towards the simultaneous treatment of multi-component data sets such as MAD, inverse-beam SAD or multi-sweep data sets related by an action of a kappa goniometer. This involves pooling, converting and re-using common information about indexing matrices at various wavelengths and for different kappa settings, and ensuring totally coherent indexing between all images sets.

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Keywords: data processing, automation, crystallographic software

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Structure of AcrB: A novel mechanism for multidrug resistance

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The AcrA-AcrB-TolC complex is the major multidrug efflux pump in Escherichia coli. The asymmetric structure of the trimeric inner-membrane component AcrB (1,2) implies functional rotation of the monomers and a peristaltic mode of drug efflux. This mechanism suggests the occurrence of conformational changes in the periplasmic pore domain through the movements of subdomains during cycling of the monomers through the different states loose (L), tight (T) and open (O). Recently, we tested this hypothesis by introducing cysteines at the interfaces of potentially moving subdomains, and obtained disulfide bond formation as quantified by alkylation of free cysteines and MALDI-TOF analysis (3). Cross-linking indeed increased susceptibility to noxious compounds , and caused inhibition of pump function which could be regained upon exposure to the reducing agent DTT.

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Structural description of the ATPase cycle of a myosin that moves backward

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Myosins are molecular motors that use the energy obtained from the hydrolysis of ATP to move along actin filaments. Among myosin family, class VI myosins are very intriguing because of their atypical motility properties. First, dimeric myosin VI is capable of taking multiple steps (processive movement) of 30-36 nm along actin filament. These steps are surprisingly very large considering that