MS4-P7 Automated refinement of low-resolution macromolecular structures using prior information

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Due to poor observation to parameter ratios at resolutions below 3Å, using additional information can improve and stabilise the refinement process. ProSMART [1] generates external restraints for protein and nucleic acid chains based on known structures of homologous molecules, idealised fragment geometries, and hydrogen bonds detected in the refined structure. These restraints are used by REFMAC5 [2] to stabilise refinement of a model. However, the optimal refinement protocol varies from case to case, and it is not always obvious how to select appropriate homologous structure(s), or other sources of prior information, for restraint generation. After running extensive tests on a large dataset of low-resolution models, we identified the best performing refinement protocols and strategies for selection of homologous structures used for restraint generation. These strategies and protocols are implemented in the Low-resolution Structure Refinement Pipeline.

The pipeline performs auto-detection of twinning and selects the optimal scaling method and solvent parameters. The pipeline can work with manually-supplied homologous structures, or run an automated BLAST search and download homologues. The pipeline tests a number of refinement protocols in order to find the best protocol for a target model, including: simple jelly-body refinement; refinement using external restraints generated from single and multiple homologues followed by relaxation with jelly-body refinement; and hydrogen bonds restraints.

We observed a strong correlation between the success of refinement using external restraints and the global RMSD between the target and homologous structures. Substantial model improvement was observed with homologues sharing as low as 75% sequence similarity. Restraints based on hydrogen bonds were found to improve refinement when no homologues were available for a particular chain. The automated pipeline improved Rfree values, geometry and Ramachandran statistics for 84% of the test cases.

The automated pipeline presented facilitates hassle-free automated selection of the optimal set of external restraints for low-resolution structure refinement and allows subsequent use of the optimal protocol in latter refinement runs.

References:

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MS4-P8 Pushing the boundaries of crystallography: Debye-Waller factor redefined

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The main purpose of crystallography is to solve and refine crystal structures based on measured diffraction data. Complex crystals structures require big datasets consisting also of weak peaks. By using synchrotron facilities and modern detectors it is possible to collect diffraction patterns with a very high dynamic range. It is, however, a big challenge to properly process the measured data. One of many corrections applied during structure refinement process is the Debye-Waller (D-W) factor correction. It compensates for the perturbations arising from thermal vibrations (phononic term) or flips (phasonic term) of atoms. The D-W factor can be also generalized to a statistical interpretation [1,2]. The general formula for D-W factor is $\exp[-k^2\sigma^2]$, where k is the scattering vector and σ is a variance of the distribution of atomic arrangement (both in physical or perpendicular space).

In our presentation we discuss the limitations of the D-W factor in terms of structure refinement and propose a way to improve the results of such analysis. We prove that the D-W factor substantially limits the range of diffraction data possible to use in a refinement process. It works correctly only for small values of the exponential in the formula above. For real crystals (including quasicrystals), satisfactorily good results are only obtained for strong reflections with intensities higher than 1% in relative scale. Peaks with intensities 10^{-4}-10^{-3} are refined rather incidentally (see e.g.[3]). This means that including weak reflections in a refinement procedure frequently makes the results worse.

We show how to improve the use of D-W factor. Our calculations are performed for a simple 1D model quasicrystal – the Fibonacci chain. Both the model choice and its low dimensionality do not affect our concluding remarks. We modeled the fluctuations in physical and perpendicular space. We claim, that redefinition of the D-W factor by either including higher-order moments of the statistical distribution or replacing the Gauss function with more appropriate functions essentially allows also weak peaks to be included in the refinement. Our results are general and can be applied for structural investigations of perfect crystals, including quasicrystals, but also any systems with defects or highly disordered.

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