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How to solve, refine, validate, and deposit difficult macromolecular structures

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The median resolution of crystallographic deposits in the PDB is 2.1 Å, making the majority of deposits well within reach of standard methods. Owing to our focus on methods development, we frequently encounter important and interesting projects that cannot be solved and refined with routine approaches. Sources of difficulties are generally related to poor macro- and micro-organization of the crystal lattice, which often results from properties of macromolecules.

In many projects, a large part of the asymmetric unit content is well-organized, so the crystals diffract well. However, the remaining parts may not be completely disorganized, and so they do contribute to diffraction, while their order may be nowhere close to data resolution. How to model, validate, and deposit such locally low-resolution deposits remains an open problem. Low-resolution may be associated with a domain that undergoes pivoting, anisotropic motion. In such cases, excluding that part from the model that is distal from the pivot point generates an awkward result that may mislead the recipient of the PDB deposit. Excluding the whole domain also may affect biological interpretation, as we will present. Additionally, as many residues are affected, so are the refinement statistics. Another option is to refine such a model with geometric, evolutionary, and NCS-constraints, permitting B-factors outside of standard ranges during refinement, e.g. above 500 Å2 [1,2]. Finally, there are also cases, e.g. a low level of order-disorder transition, when the most appropriate course of action is to ignore the disordered parts [3].

We will present the several examples of such problems on systems ranging from 1.9 A to 3.9 A where we applied multiple lines of validation to assure that the final models were correct. In some cases, we had additional a posteriori validation by higher-resolution structures of the fragments.

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2. Leung, D.W. et al. (2015). Cell Rep. 11, 376-389.
3. 5V10.pdb

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