Poster Presentations

[MS04-P01] Prediction of alternative and disordered residues by unrestrained refinement. Oleg V. Sobolev, Vladimir Y. Lunin,

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Unrestrained reciprocal space refinement is stable for the vast majority of atoms when working at atomic resolution. Nevertheless geometrical restraints should be held in refinement for residues that are present in several (alternative) conformations in the crystal used for the X-ray experiment; in the other case such residues deteriorate significantly. We have shown that on the contrary, large distortion of a residue in unrestrained refinement may hint the presence of alternative conformations (ACs) for this residue [1]. To get these hints in a routine way we have implemented two procedures that analyze amplitudes of atomic shifts resulting from several cycles of unrestrained refinement as computer programs [2]. A simple diagram plotting the values of atomic shifts against the residue number may give an idea of the crystallographic order of different parts of the structure at qualitative level. The second approach presents an automatic decision-making procedure that classifies each residue as 'single conformation' or 'alternative conformations'. The decision is made either by comparing the observed atomic shifts against a predetermined threshold, or by comparing probabilities of these shifts for residues possessing single and multiple conformations.
The prediction quality of suggested procedures and parameters of decision-making procedures were derived using author's allocation of ACs deposited in PDB. Nevertheless, these allocations may not always precisely represent the actual state of a residue. The main reason for the misleading allocations is ill-defined areas of electron density maps around poorly ordered residues. Such residues are left in SCs in the model despite they are obviously present in multiple conformations in the crystal. When better data will be available some of such poorly ordered residues may be modeled in ACs. To illustrate this observation, we selected three accurate structures of lyzocime (129 residues) from PDB: 4lzt (0.95Å, room temperature), 3lzt (0.93Å, cryo temperature), 2vb1 (0.65Å, cryo temperature) crystallized in the same space group P1. 14, 28 and 49 residues were modeled with alternative conformations in 4lzt, 3lzt and 2vb1 respectively. One residue in 4lzt and seven residues in 3lzt modeled in ACs were left in single conformation in 2vb1 model. We applied our approach to find residues in ACs for 4lzt structure and analyzed the magnitudes of atomic shifts with our programs. The programs detected 53 residues as primary candidates to be modeled in alternative conformations. 30 residues detected as AC by our programs were not included in 4zlt model in ACs but were modeled with ACs in 2vb1 model. For 8 residues suggested by programs, ACs were not included even in 2vb1 model. The most of them are terminal residues with poor electron density which does not provide positions of alternative conformations.
The presented study shows that analysis of atomic shifts in unrestrained refinement may reveal poorly ordered residues that should be checked first with electron density maps for modeling in ACs. This analysis is also sensitive to disordered residues which may be modeled in ACs when better ordered crystals will be available.

The programs and documentation may be downloaded from the site:
www.impb.ru/lmc/programs/ac_prediction/.
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[1] Sobolev O.V., Lunin V.Y. (2012). *Acta Cryst.* D68. 1118-1127.
[2] Sobolev O.V. (2013). J. *Appl. Crys*t. 46. 554-559.

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