



KURS PROTEINOVÉ KRYSTALOGRAFIE

CRYSTALLIZATION METHODS USED IN PROTEIN CRYSTALLOGENESIS

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Finding suitable crystallization conditions is the main problem to solve a protein structure by X-ray diffraction techniques.

In this lecture:

classical crystallization techniques based on evaporation used for screening and optimization of crystallization conditions utilizing the screening upon previously successful chemical cocktails,

advanced counter-diffusion technique that allows the screening for crystallization conditions in a wide range of supersaturation while suppressing concentration, of protein and precipitant,

cross-crystallization procedure based on using additives to modify crystal morphology and to improve diffraction quality,

will be discussed.

Literature:

Ivana Tomčová and Ivana Kutá Smatanová: Copper co-crystallization and divalent metal salts cross-influence effect – a new optimisation tool improving crystal morphology and diffraction quality. *Journal of Crystal Growth*, accepted for publication (2007).

Ivana Tomčová and Ivana Kutá Smatanová: Cross-crystallization as a new optimization tool of crystallization procedures. *Materials Structure* 14, 1, 3-5 (2007).

Ivana Kutá Smatanová, José A. Gavira, Pavlína Řezáčová, František Vácha, and Juan M. García-Ruiz: New techniques for membrane protein crystallization tested on photosystem II core complex of *Pisum sativum*. *Photosynthesis Research* 90 (3), 255-259 (2006).

Ivana Tomčová, Rui Miguel Mamede Branca, Gabriella Bodó, Csaba Bagyinka, and Ivana Kutá Smatanová: Cross-crystallization and preliminary diffraction analysis of a novel di-heme cytochrome *c₄*. *Acta Cryst.* F62, 820-824 (2006).

Julie Wolfova, Rita Grandori, Erika Kozma, Neal Chatterjee, Jannette Carey and Ivana Kuta Smatanova: Crystallization of the flavoprotein WrbA optimized by using additives and gels. *Journal of Crystal Growth* 284, 3-4, 502-505 (2005).

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DEPOSITION OF MACROMOLECULAR STRUCTURES TO THE PROTEIN DATA BANK (PDB)

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Most grant agencies and virtually all journals require that the result of crystallographic or solution NMR analysis are deposited with a public database. In case of macromolecular structures, it is the Protein Data Bank ([1], PDB, <http://www.pdb.org/>) or the Nucleic acid Database ([2], NDB, <http://ndbserver.rutgers.edu>). Everyone involved in structure determination should keep in mind that structures that have been nurtured in laboratories for months and in some cases for years, will not be viewed in light of notebooks, log files from data processing and refinement, neither from endless coffee discussions in the laboratory but solely by their representation in the PDB. The deposition process therefore deserves attention and should be viewed as an important part of structure determination. The work-

shop will present the tools developed by the RCSB PDB that assist and simplify the deposition.

The main deposition tool is AdIt, deposition and validation tool, <http://deposit.rcsb.org/>. It is a web-based mmCIF editor. To deposit a structure, the user uploads the relevant coordinate and experimental data files and then adds any additional information. Each structure should be validated before deposition. Coordinates should be checked for format consistency and for quality of valence geometry using the Validation server (<http://deposit.pdb.org/validate/>). Web server <http://pdb-extract.rcsb.org/auto-check/> allows non-trivial checking of coordinates versus x-ray diffraction data („structure factors“) using programs SFCheck, REFMAC, and CNS. Correctly formatted coordinates as well as collection and refinement statistics should be pro-

duced by the `pdb_extract` tool ([1], <http://pdb-extract.rcsb.org/>) that allows integration of refinement logs of most major refinement programs into PDB and/or mmCIF format and significantly thus simplifies the deposition. Identity of ligands present in the to-be-deposited structure should be verified using the `ligand` tool, currently at the web for „Ligand Depot“ (<http://ligand-depot.rcsb.org/>) that allows you to determine whether your ligands are correctly labeled, whether the right atom names were used, and whether these ligands are possibly new to the PDB.

All the mentioned web pages have available extensive tutorials, many steps have context-sensitive help and example pages and most of them are available as downloadable executable files as well as source codes.

The workshop will show deposition process using example files, possibly from participants.

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