

## MS58.P06

*Acta Cryst.* (2011) A67, C593**Incorporation of the quantum chemical package DivCon into the PHENIX suite**

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X-ray crystallography is the primarily technique used to reveal the three-dimensional structure of protein complexes that play a critical role in Structure Based Drug Design. Because of the low ratio of observed data to refined parameters, macromolecular crystallographic refinement at moderate and low resolution relies heavily on the set of known amino acid geometric parameters that ensures the correct stereochemistry of the model. Available programs for macromolecular refinement such as REFMAC, PHENIX or SHELX use simple harmonic oscillator functions to introduce stereochemistry restraints and commonly do not account for electrostatic interactions in the system. This approach inevitably masks important structural details that are often crucial to the understanding of ligand binding within the active site of the protein. To overcome these limitations and achieve a much more realistic description of the protein-ligand geometry, we replaced these stereochemistry restraints with the energy functional derived from quantum-mechanical (QM) treatment. This treatment has been demonstrated with the successful integration of the commercial DivCon ToolKit developed by QuantumBio with the popular Python-based crystallographic package PHENIX. DivCon employs semiempirical QM methods such as AM1, PM3 or PM6 and is based on the divide-and-conquer approach to evaluate the density matrix allowing linear-scaling of the QM problem. As a result, DivCon dramatically decreases the computation costs traditionally associated with QM-based methods, making the application of quantum chemistry for large protein systems feasible.

We proposed a novel protocol to incorporate QM gradients and energy targets into individual (“XYZ”) coordinate refinement step in PHENIX without altering the other refinement stages such as the bulk solvent correction or temperature factor refinement. Furthermore, a user has a choice to use DivCon methods either for the whole structure or a selected region - a ligand and protein active site residues, for example. Based on five test protein structures downloaded from Protein Data Bank (PDB) we report the detailed comparison of the conventional and QM driven refinements. Our preliminary results indicate that incorporation of the QM function not only improves the local geometry but also reduce the R/Rfree factors. For example, the re-refinement of a 17-residue short protein at 2 Å resolution (PDB ID 1S9Z) using the PHENIX/DivCon package indicates a number of significant structural improvements as compared to the conventional PHENIX refinement. Notably, PHENIX/DivCon compared to PHENIX alone improved the peptide bond geometry for the non-standard N terminus residue of that protein and the DivCon driven refinement accurately represents the stereochemistry in this region. Furthermore, the QM approach results in more reasonable H-bond network throughout the protein molecule.

**Keywords:** DivCon, phenix, refinement

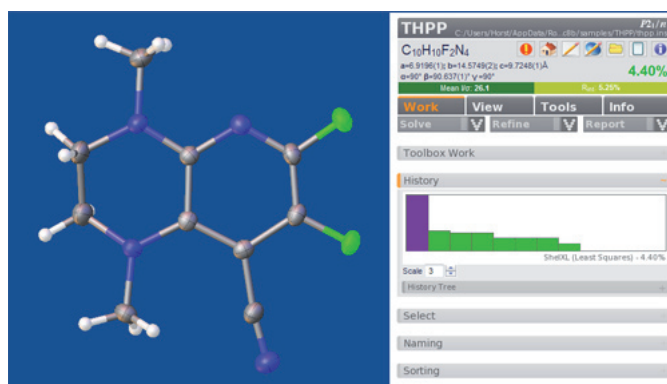
## MS58.P07

*Acta Cryst.* (2011) A67, C593**Olex2 – A complete package for molecular crystallography**

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Olex2 [1] has become established in the community of small-molecule crystallographers as an easy-to-use unified package that provides tools needed for day-to-day analyses of small molecule structures. There is a rapidly growing number of installations of the software world-wide and our core paper [2] has attracted a significant number of citations since 2009.



**Structure Solution** is achieved by our own charge-flipping implementation, *olex2.solve*, based on E<sup>2</sup>.

**Structure Refinement** can be carried out with *olex2.refine*. The refinement engine is based on the cctbx and provides all the functionality required for a meaningful structure refinement. A general system allows the implementation of any constraints, which has been used to provide all ShelXL constraints.

The solution and refinement programs are based on the small-molecule toolbox (*smtbx*), that our group has contributed to the family of tools available in the *cctbx* [3].

**Structure Analysis** tools covering most requirements are an integral part – growing, packing, geometric measurements, void, molecular and solvent accessible volume calculation,  $\pi$ - $\pi$  analysis and many more.

**Structure Publication** is made easy. Complete and correct CIFs result automatically, the generation of reports is easy and images – bitmaps or ORTEP-style drawings – can be generated with minimum effort.

Olex2 is Open Source under the BSD Licence and available free of charge from [1] for academic users for Windows, Linux and MacOSX.

[1] [www.olex2.org](http://www.olex2.org) [2] O.V. Dolomanov, L.J. Bourhis, R.J. Gildea, J.A.K. Howard, H. Puschmann, Olex2, *J. Appl. Cryst.* **2009**, *42*, 339-341. [3] R.W. Grosse-Kunstleve et al, <http://cctbx.sourceforge.net/>

**Keywords:** refinement, molecular, software

## MS58.P08

*Acta Cryst.* (2011) A67, C593-C594**Full matrix refinement with the small molecule toolbox**

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The Computational Crystallography Toolbox (cctbx) [1] opened a new era in crystallographic computing by providing a free, open and

comprehensive implementation of the fundamentals of crystallography (symmetries, Fourier, scattering, etc). As the foundation of the macromolecular suite PHENIX, it has a certain connotation which is undeserved since the algorithms and data structures it features are correct for any crystal structure.

As part of the EPSRC grant "Age Concern" we developed a companion library, the Small Molecule Toolbox (smtbx). It shares the same philosophy as the cctbx: it is designed to make the writing of short scripts easy as well as to make it possible to build or to integrate it into large programs. It provides tools covering the whole workflow of small molecule work but we will focus on refinement in this talk.

The smtbx provides full matrix least-squares, restraints on bond lengths, angles, or dihedral angles, and special position constraints as well as the wealth of geometrical constraints available in ShelX; the both of merohedral and non-merohedral twin refinement; a solvent disorder modelling similar to the SQUEEZE procedure in PLATON.

More importantly, its modular and open design ease the addition of new features. We will present one such new tool to model water molecules.

[1] R. Grosse-Kunstleve et al, <http://cctbx.sourceforge.net/>

**Keywords:** refinement, full matrix, constraints

## MS58.P09

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### On the refinement of routine single crystal X-ray data only to mimic single crystal neutron structural results

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In the case of organic compounds single crystal neutron diffraction is a source of reliable structural data particularly hydrogen atom positions and their ADPs. In consequence, neutron geometry of organic molecules is usually more reliable than single crystal X-ray diffraction structural data, although no doubt this is the X-ray diffraction technique which is by far the most popular among crystallographers to acquire structural information.

One can ask then a question whether it is possible to refine single crystal X-ray diffraction data only in such a way as to mimic the geometry of molecules obtained from single crystal neutron diffraction experiments.

In this contribution will present results of our analysis focused on comparison of structural neutron and X-ray results obtained for a series of five crystals of model compounds of increasing complexity and quality of data.

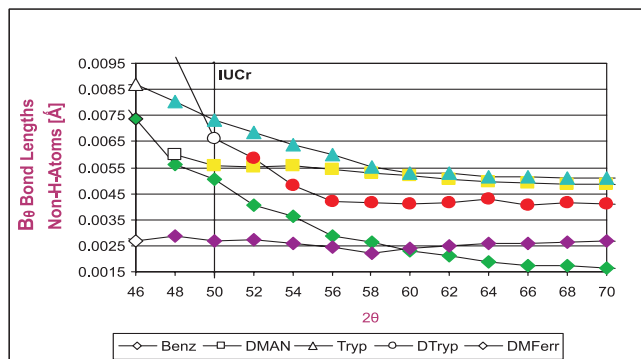


Fig. 1. Average differences between the neutron and X-ray bond lengths for the non-H-atoms obtained for a series of model compounds as a function of diffraction  $2\theta$  angle.

For each crystal, we have performed series of refinements as a function of resolution ( $\sin \theta/\lambda$ , in fact as a function of diffraction  $2\theta$  angle) using the neutron structural results as the reference ones. Will present a number of dependences of different parameters characterising the quality of X-ray data sets and average differences between particular neutron and X-ray structural parameters on  $2\theta$  diffraction angle. The results obtained influence understanding of benchmarks commonly accepted by IUCr and used in different checkcif programs, in particular  $2\theta$  limit equal to  $50^\circ$  for the  $\text{MoK}\alpha$  X-ray radiation. One example of such a dependence is shown in figure below.

**Keywords:** neutron, X-ray, refinement

## MS58.P10

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### On a method for the absolute scaling of refined atomic B factors

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The comparison of crystallographic models representing different functional and/or physical states of the same protein might seem easy if big conformational differences are detected by simple visual inspection. The precision of the models must be immediately considered if subtle structural changes are at play or if the aim is to evaluate small internal changes that occur in combination with for example large scale rigid-body changes. While the estimation of the positional uncertainty of crystallographic models has been extensively studied and a few methods have been developed to perform error-inclusive coordinate comparisons, the comparison of isotropic atomic B factors ( $B_i$ ) has received less attention. One problem is to estimate standard uncertainties for refined  $B_i$ , but in addition, it is known that direct quantitative comparisons of  $B_i$  are flawed by several sources of model-specific variations (such as refinement strategies, data resolution, etc) ultimately contributing to a general scaling problem (inaccuracy). Precise and accurate comparison of  $B_i$  among several (frequently many) models would represent a rich source of information about dynamics and plasticity, often neglected in crystallographic approaches, despite the general acceptance of using  $B_i$  as a critical parameter in model refinement. The development of new methods to bring refined B factors to a common absolute scale would thus represent a genuine contribution, especially relevant to tackle the problem of "protein allostery without conformational change" (i.e. without a change in the shape: conformational entropy modulation). In this work we propose a method to compare atomic B factors, based on Cruickshank's approach to predict atomic positional standard uncertainties (psu's) in crystallographic models [1]. The method we are now proposing uses Cruickshank's psu's to build a correction index that, once applied to individual refined atomic B factors, generates a set of scaled B factors to be used for comparison purposes. This approach assumes that the psu's integrate all sources of position uncertainty: dynamic (temperature-dependent), static and model error. Among other interesting applications, direct comparison of room and cryogenic temperature models are possible. Different sets of protein models consisting of apo and complexed forms revealed a previously overlooked inverse relationship between  $B_{\text{avg}}$  and  $R_{\text{free}}$ , warning about the need for adequate and complete model refinement strategies to ensure accurate structural comparisons. We show evidence supporting that the Wilson B factor value can act as a universal attractor leading to inaccurate models. As a practical example we have used