

## MS45-O3 SEQUENCE SLIDER: a multi sequence evaluator and its application in venomics

Rafael J. Borges<sup>1,2</sup>, Massimo Sammito<sup>1</sup>, Claudia Millán<sup>1</sup>, Marcos R.M. Fontes<sup>2</sup>, Isabel Usón<sup>1,3</sup>

1. Dept. of Structural Molecular, Institute of Molecular Biology of Barcelona (IBMB), Spanish National Research Council (CSIC), Barcelona, Spain

2. Dept. of Physics and Biophysics, Biosciences Institute (IBB), São Paulo State University (UNESP), Botucatu, São Paulo, Brazil

3. Dept. of Structural Biology, Catalan Institution for Research and Advanced Studies (ICREA), Barcelona, Spain

email: rjborges@ibb.unesp.br

Structural biology has been an invaluable tool in the biomedical field of venomics with drug design, comprehension of physiological mechanisms and improvement of antivenom therapy (Calvete et al., 2009). Venom extraction is a common procedure as it is stored in an accessible gland. Thus, often, snake toxins are directly obtained from natural venom purification, rather than through recombinant protein production. Characterized by one of the most rapid evolutionary divergence and variability in any category of proteins, venoms are composed by a cocktail of toxins (Calvete et al., 2009). With such complex composition, it is a challenge to obtain high purity toxins, since physico-chemical properties may be shared among different proteins. Thus, the obtained samples from final steps of purification can be a mixture of isoforms and/or different protein. In such cases, the sequence may not be clearly determined by mass spectrometry and even after protein isolation through crystallization, determining side chain composition in the crystallographic model may not be trivial. SEQUENCE SLIDER aims to address such scenario integrating other sources of experimental data. Starting from a partial or complete model assigning and evaluating different sequence possibilities against diffraction data. The side chains are modeled combining hypotheses assembled with SCWRL4 (Krivov et al., 2009) and/or COOT (Emsley et al., 2010) and refinement through REFMAC (Murshudov et al., 2011) and/or BUSTER (Bricogne et al., 2011). The varied sequences are generated using a probability distribution based on experimental results of mass spectrometer or on phylogenetic statistics. Correct model discrimination is evaluated through proposition of a Figure of Merit integrating global and local crystallographic indicators, energy and complementary experimental knowledge. SEQUENCE SLIDER is being applied to determine unsolved snake venom toxin crystallography datasets and revisit toxins deposited in the protein databank.

### References

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**Keywords:** Venomics, Crystallography, protein sequencing, refinement, modeling, phasing, ARCIMBOLDO, mass spectrometry

## MS45-O4 DSR – Enhanced modelling and refinement of disordered structures with SHELXL

Daniel Kratzert<sup>1</sup>, Ingo Krossing<sup>1</sup>

1. Albert-Ludwigs-Universität Freiburg

email: dkratzert@gmx.de

One of the remaining challenges in single-crystal structure refinement is the proper description of disorder in crystal structures. DSR performs semi-automatic modelling of disordered moieties in SHELXL<sup>[2]</sup>. It contains a database that includes molecular fragments and their corresponding stereochemical restraints and a fitting procedure to place these fragments on the desired position in the unit cell. The program is also suitable for speeding up model building of well-ordered crystal structures.

Writing a special DSR command into the SHELXL .res file of the target structure instructs DSR on where to place and how to orient a molecular fragment from the fragment database in the unit cell (Figure 1). In addition, it is possible to define the occupancy, residue or part number of the fitting fragment. The fragment can be inverted, e.g. to fit the other conformer of twisted tetrahydrofuran.

Molecular fragments can be either imported directly from the GRADE server of Globalphasing Ltd.<sup>[2]</sup>, from existing crystal structures or from ab initio calculations. DSR offers several more options available to make disorder modelling a convenient process.

[1] G. M. Sheldrick, *Acta Cryst.* **2015**, *C71*, 3–8.

[2] Grade Web Server, *Global Phasing Ltd.* <http://grade.globalphasing.org>.

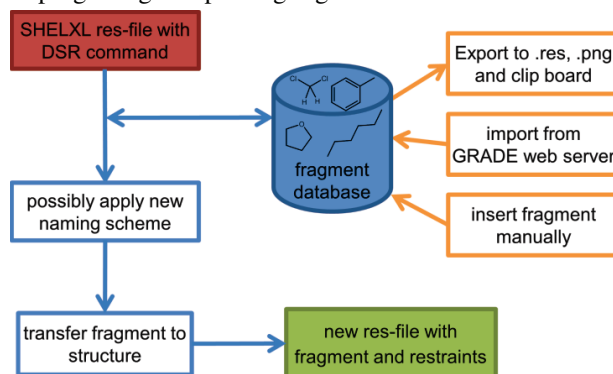


Figure 1. General work flow of the DSR program.

**Keywords:** X-ray structure, refinement, disorder, SHELXL, molecular fragments.