Instruct/CIISB course on fragment screening using crystallography laboratory equipment

April 5-6, 2018 (arrival in Vestec April 4, departure from Vestec April 7 after breakfast)

Venue: Centre of Molecular Structure, IBT CAS v.v.i., BIOCEV, Vestec, Prague-West, Czech Republic

Accommodation for the participants to the practical (hands-on) sessions:

shared rooms (2 people), Hotel u Krbu, Vídeňská 360, 252 42 Vestec, Czech Republic

Aims of the course:

Fragment-screening (FS) is a popular approach to identify the binding of small organic molecules (fragments) to protein targets. This may be done to elucidate the function of a protein or to identify target-binding chemical structures, which then may be developed into inhibitors in a process called lead compound discovery.

Fragment screening campaigns require the availability of a library (or libraries) of low-molecular weight compounds, several of which are accessible to the public.

A campaign normally takes place in two stages: an initial stage of pre-screening using biochemical or biophysical methods, such as thermal shift assay or Surface Plasmon Resonance.

Afterwards, the most suitable method for the second stage of fragment screening is undoubtedly X-ray crystallography, using crystals of the target macromolecule. For convenience, in-situ screening can be performed, using crystals in 96-well trays. Until recently, such screenings were normally done at synchrotron radiation sources, well equipped for in-situ diffraction.

Recently, the availability of high-flux laboratory X-ray sources (based on liquid Gallium alloy jet technology for the anode material) together with the availability of commercial stages for in-situ diffraction means that the second stage of fragment screening can be performed in well-equipped macromolecular crystallography laboratories.

The first aim of the workshop is to introduce fragment-screening practices to the audience.
The second aim is to demonstrate, in a practical way, the feasibility of such laboratory fragment screening campaigns. For this reason, the workshop will take place at the Centre of Molecular Structure (CMS), Institute of Biotechnology CAS v.v.i., BIOCEV in Vestec, which is one of the components of CIISB and is one of the Instruct CZ sites.

For X-ray diffraction, the CMS is equipped with a Bruker D8Venture diffractometer with a liquid Gallium jet anode Excillum source, Photon II detector and an ISX stage for in-situ diffraction. For relevant biophysical measurements, the Centre offers access to microscale thermophoresis (MST), to differential scanning fluorescence (DSF), and to Surface Plamon Resonance (SPR).

Expected impact for (young) researchers:

The workshop will allow participants for the hands-on sessions (16), with some prior knowledge and experience in practical macromolecular crystallography, to access the biophysical measurements (microscale thermophoresis and/or surface plasmon resonance) and X-ray diffraction sections of the Instruct CZ / CIISB CMS location. The workshop will allow them to acquire knowledge and practical experience on how to apply fragment-screening techniques to a real case. Depending on the proposals made by the participants, one or two of the participants' project may be selected to be used for demonstrations (one sample suitable for biophysical measurements; a sufficient number of one type of macromolecular crystals – both brought by the selected participants, to be arranged in advance).

Opportunity for participants to bring their own samples: Yes (see above), must be arranged in advance

Contact: F. Vellieux (frederic.vellieux@ibt.cas.cz). To apply, send a letter by email indicating how you would benefit from attendance. In case of junior scientists, a letter of confirmation from your supervisor is also requested. Please also indicate if you would like to bring a poster.

Registration fee: none – the registered participants (16) are expected to cover the costs of travel to and from Vestec (arrival in Vestec on April 4 evening, departure from Vestec on April 7 morning). The registration for the 16 selected participants (for the practical sessions) covers all costs from the first night in Hotel u Krbu to the final night (departure after breakfast on April 7). Note that accommodation plus catering costs will be charged in case of "no show".

Course content:

Introductory talks (Biocev auditorium, April 5 morning session, **everyone can attend** without registration)

Opening and welcome words - CIISB (Jan Dohnálek, IBT) **10'** (09:00 – 09:10)

A. Heine (IPC Marburg): Crystallographic fragment detection:
screening results for selected targets using a 96-fragments library **40'** (09:10 - 09:50)

B. Schneider (IBT): Water as the smallest fragment **20'** (09:50 – 10:10)

T. Koval (IBT): Experiences with biophysical investigations on ligand binding **20'** (10:10 – 10:30)

P. Pompach (CMS): Proteins/small molecules interactions monitoring by HDX **20'** (10:30 – 10:50)

Coffee break and group photo **30'** (10:50 – 11:20)

M. Weiss (Bessy II): Fragment-screening by crystallography **40'** (11:20 – 12:00)

A. Gutmanas (PDBe): The PDB, databases and archiving the structures of fragments **40'** (12:00 – 12:40)

John Darby (York): Fragment linking and the biophysical follow-up process **40'** (12:40 – 13:20)

Lunch for the registered course participants and speakers, posters and discussion

Practical sessions of fragment screening at CMS (workshop part of the course, for 16 participants):

Going from basic biophysical characterization of the samples all the way to in-situ diffraction testing and then single crystal data collection using the D8Venture ISX stage. This practical part will be divided into three sessions:

1) pre-screening using biophysical measurements (½ day, Apr 5 afternoon)
and (½ day, Apr 6 morning);

2) dispensing target protein crystals to a fragment library (½ day, Apr 5 afternoon)
and (½ day, Apr 6 morning);

3) *in situ* testing of crystal diffraction; single crystal data collection and evaluation of the results (½ day, Apr 6 afternoon).

Course dinner: Vila Lanna, V Sadech 1, Prague (April 6, evening)

Number of participants: **16**. Applications originating from laboratories abroad are especially welcome.

Attendance will be virtually unlimited for the talks (taking place in the Biocev auditorium on April 5, morning – no formal registration is requested from people who will attend the talks nor fee requested).

Number of registered participants for the practical sessions at the CMS: 16, as indicated above.

Course tutors / lecturers:

Drs Andreas Heine (Institute of Pharmaceutical Chemistry, Philipps-University Marburg), John Darby (Department of Chemistry, University of York), Aleksandras Gutmanas (European Bioinformatics Institute), Manfred Weiss (Helmholz Zentrum Berlin & Bessy II), Tatsiana Charnavets (CMS, IBT, BIOCEV), Tomas Koval (IBT, Biocev) Jiri Pavlicek (CMS, IBT, BIOCEV), Petr Pompach (CMS, IBT, BIOCEV), Bohdan Schneider (IBT, BIOCEV).