Small Angle Scattering Platform for Structural Biology

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OUTLINE:
- SAXS/SANS in Grenoble: new SAS platform of CISB
- Conversion of ID14-EH3 from MX to bio-SAXS
• Instruments available to European biological community
• Joint proposals SAXS/SANS
• Expertise
  – use of instruments
  – experimental protocols
  – data interpretation

**ILL:** Peter Timmins, P. Callow, R. May,
**IVMS:** M. Jamin,
**EMBL:** J. Marquez,
**IBS:** E. Pebay-Peyroula, M. Blackledge, F. Gabel,
**ESRF:** S. Larsen, C. Ferrero, P. Pernot, D. Spruce
SMALL ANGLE SCATTERING

= a technique for studying structure and association at low resolution in solution under normal biochemical conditions

**Information from SAS:**
- model independent parameters \( R_g, I(0) \)
- \textit{ab initio} shape determination
- rigid body modelling

- molecular shape
- molecular interactions
- kinetics

- complementarity of SAXS and SANS

<table>
<thead>
<tr>
<th></th>
<th>SAXS</th>
<th>SANS</th>
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<tbody>
<tr>
<td>volume</td>
<td>small &lt; 50 ml</td>
<td>larger ~ 300 ml</td>
</tr>
<tr>
<td>concentration</td>
<td>&gt; 0.1 mg/ml</td>
<td>&gt; 1 mg/ml</td>
</tr>
<tr>
<td>measuring time</td>
<td>short ~ s</td>
<td>longer ~ m+h</td>
</tr>
<tr>
<td>radiation damage</td>
<td>yes</td>
<td>no</td>
</tr>
<tr>
<td>contrast variation</td>
<td>no</td>
<td>yes</td>
</tr>
<tr>
<td>sensitive to salts, denaturants</td>
<td>yes</td>
<td>no</td>
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INSTRUMENTS FOR SAS

- SAXS – ESRF
  - ID2
  - ID14-3 conversion: budget ESRF, scientist EMBL

- SANS – ILL
  - D11
  - D22
  - D33 project

- Instruments available via proposals to ESRF and ILL
ID14-EH3 conversion to bio-SAXS

GOAL = investigation of biological macromolecular complexes in solution

$q_{\text{min}}$ and $q_{\text{max}}$ defined by the experimental setup:
- $\lambda = 0.931\text{Å}$, $(E = 13.3 \text{ keV})$
- detector diameter $\sim 20$ cm,
- sample-to-detector distance $D: 1-3 \text{ m}$
- beam stop size $\sim 2 \text{ mm}$

| $D$ | $q_{\text{min}}$ | $d_{\text{max}}$ | $q_{\text{max}}$ | $d_{\text{min}}$
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<tr>
<td>$1 \text{ m}$</td>
<td>$0.013 \text{ Å}^{-1}$</td>
<td>$47 \text{ nm}$</td>
<td>$1.34 \text{ Å}^{-1}$</td>
<td>$4.68 \text{ Å}$</td>
</tr>
<tr>
<td>$3 \text{ m}$</td>
<td>$4.3 \times 10^{-3} \text{ Å}^{-1}$</td>
<td>$140 \text{ nm}$</td>
<td>$0.45 \text{ Å}^{-1}$</td>
<td>$14 \text{ Å}$</td>
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REQUIRED MODIFICATIONS

q resolution limited by beam divergence, beam size, parasitic scattering, detector PSF and dynamic range, ...
OPTIC HUTCH

- beam defining slits after the mirror
- fluorescent screen after the slits
- attenuators before Ge crystal

DONE during October shutdown
MINIHUTCH in mezzanine

~ 4 m from the sample

Status:
- all components ordered
- safety approval obtained
- installation during the winter shutdown
EXPERIMENTAL HUTCH

January-March 2008
- commissioning of new electronics of Optic hutch, minihutch equipments and feasibility SAXS tests
- equiped as it is, however without Be-window, with new table to hold detector
~ 2.6 m from the sample (small cell on microdiff), flight tube, motorized beamstop
**EXPERIMENTAL HUTCH**

*from March 2008:* installation of new equipments and commissioning

**FLIGHT TUBE** with modulable length, height
- with large Kapton window
- motorized beam stop with an incorporated diode close to detector

**GUARD SLITS** in vacuum as close possible to the sample

**SAMPLE cell**

**Kapton window**

Fluorescent screen and diode near to sample position

**X-rays**

**Status:**
- decisions pending…
Present on the beamline: Q4R ADSC
- $2 \times 2$ array with no more than 400 $\mu$m slit between detecting areas
- $2304 \times 2304$ pixels, $82 \, \mu m^2$ each
- active area: $188 \times 188 \, mm^2$
- readout time: lowest noise $\sim 9 \, s$
  high speed $\sim 3 \, s$
- 16 bits
Fiber optically coupled (taper) Frelon CCD based on **Kodak KAF-4320** image sensor
- Active area: $10 \text{ cm} \times 10 \text{ cm}$
- Full dynamic range: 16 bit (14.5 bit above the noise)
- Spatial resolution: $80 \mu\text{m}$ (49 $\mu\text{m}$ pixel)
- Full frame rate of 3 frames/sec ($2048 \times 2048$)
- Detector translation table to cover required q-range

ID02 installation
Bruker AXS
VÅNTEC-2000

*Gas-filled detector bought by Detector pool*

14 cm x 14 cm active area
100 msec Snapshot
spatial resolution = 70 µm
local and global count rate of >800,000
high dynamic range of >10⁷
high area uniformity

**Inert counting gas**
no maintenance required

**Radiation hard**
can withstand primary beam

all advantages of a sealed, gaseous, photon-counting detector with real-time mode
PILATUS

- high dynamic range: > 1:1 000 000 (20bits)/pixel/image
- readout time ~ 3 ms
- no electronic noise
- point spread function = 1 pixel
- electronically gateable: no mechanical shutter needed
- pixel size: 172 x 172 µm²
- single module: 195 x 487 pixels
- active area single module: 33.5 x 83.8 mm²
- counting rate: > 2 x 10⁶/s/pixel
- framing rate: 200 Hz
- energy range: 3 – 30 keV

‘Stripe’ (5 modules, 500K) bought by the bio-SAXS beamline at EMBL Hamburg borrow to the ESRF from January to September 2008
Timetable 2007-8

ID14-EH3 as MX operation

no perturbations for others ID14 end-stations

Commissioning as SAXS with user volunteers
ID14-EH3 conversion to bio-SAXS

people involved

Cooperation with EMBL Hamburg: D. Svergun, M. Rössle
- actual bio-SAXS beamline at Doris, future at PETRA III
- PILATUS, ‘mini’ Sample Changer, data analysis software, etc.
Via EMBL Grenoble: F. Cipriani, A. McCarthy

Advice from ID02 and BM26 ESRF: N. Theyencheri, P. Bösecke, and W. Bras

MX ‘realisation team’: P. Theveneau, D. Spruce, T. Mairs, D. Nurizzo

Still waiting for EMBL expert…: NN