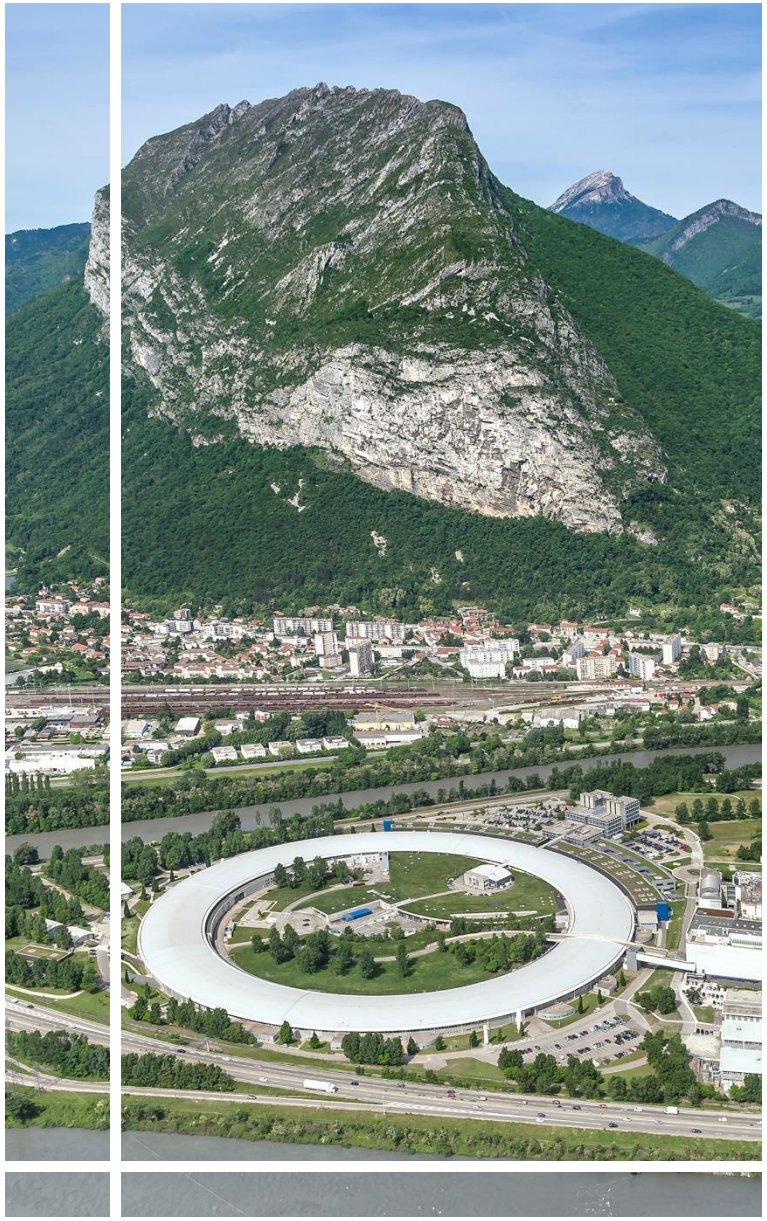
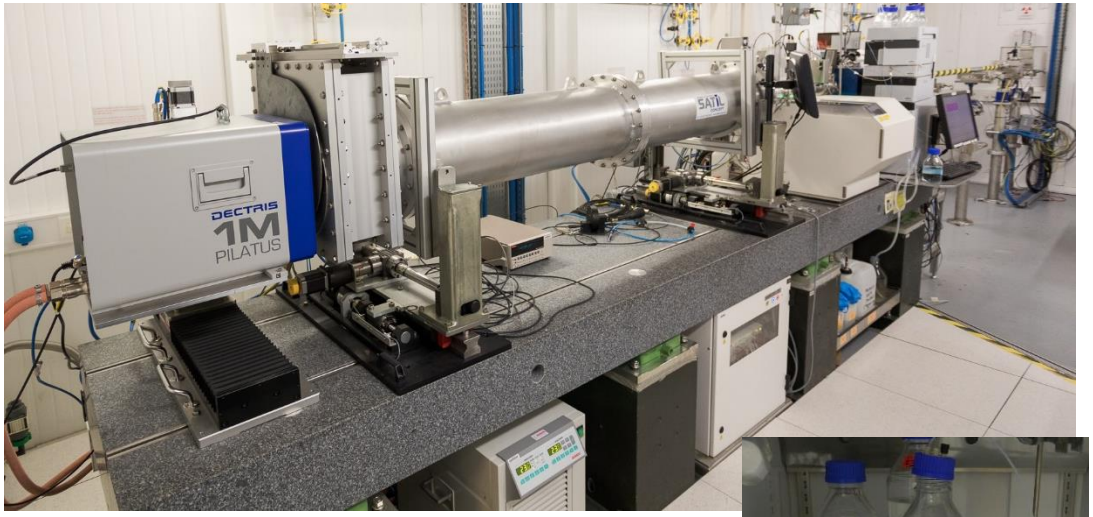
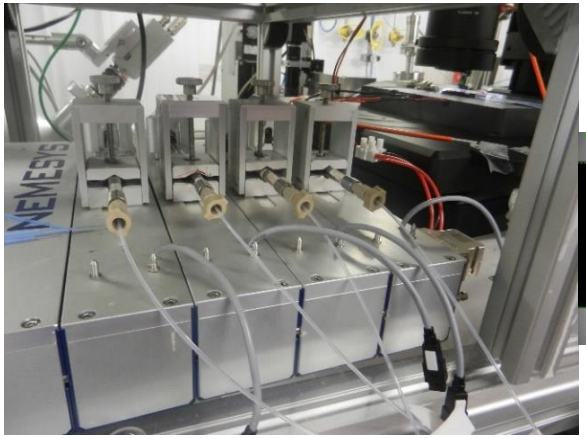
BM29: BIOSAXS BEAMLINE STATUS AND NEWS

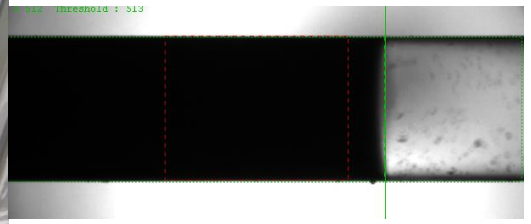


BM29



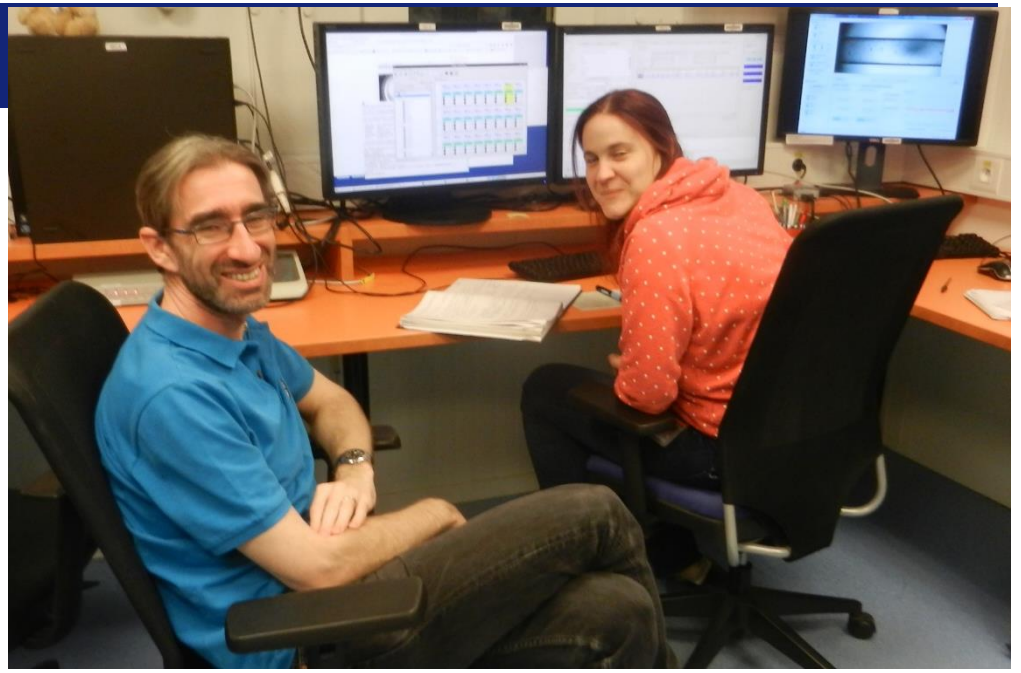
Petra Pernot
ESRF
5th February 2018
BAG Meeting





The European Synchrotron | ESRF

BIOSAXS LCS



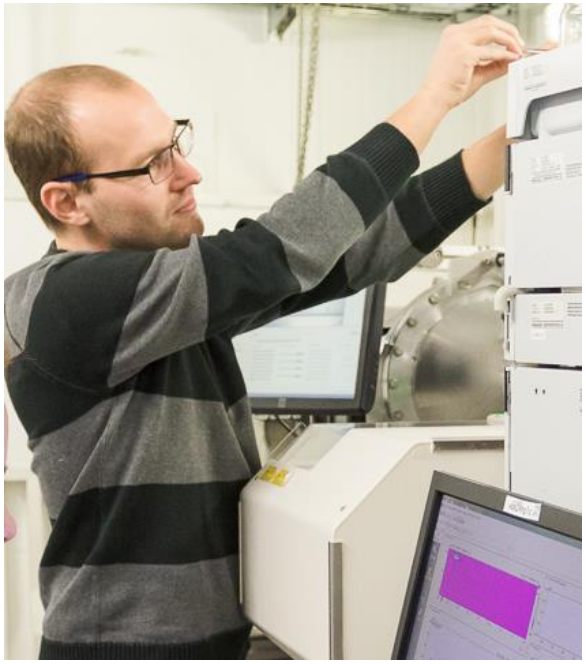
Mark Tully: ESRF scientist
- started March 2017

Martha Brennich: EMBL scientist
- formerly ESRF PostDoc and
Junior scientist

Gabriele Giachin

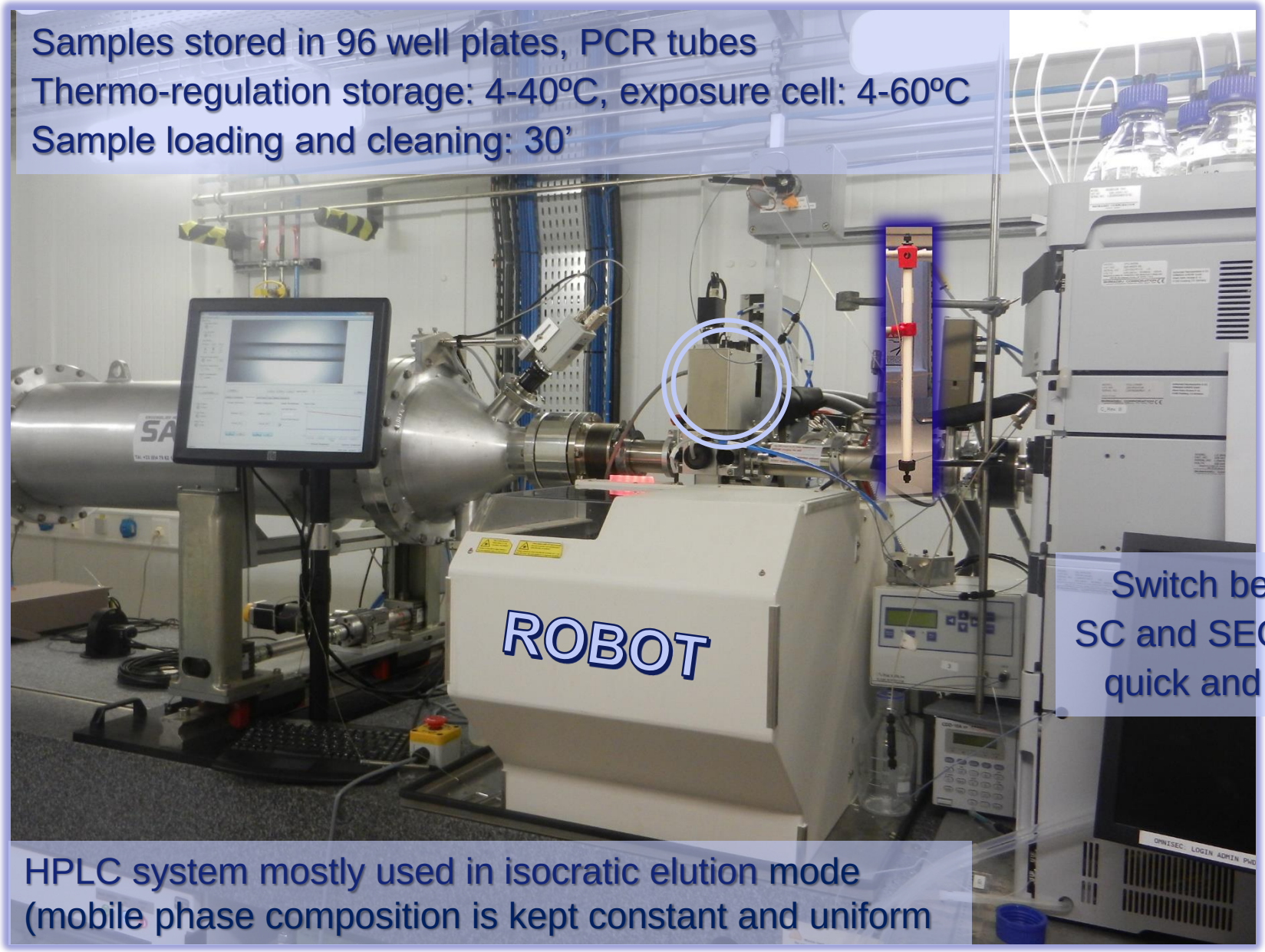
and Bart van Laer

ESRF
Junior scientists



BM29 MAIN HORSES: SAMPLE CHANGER + HPLC SYSTEM

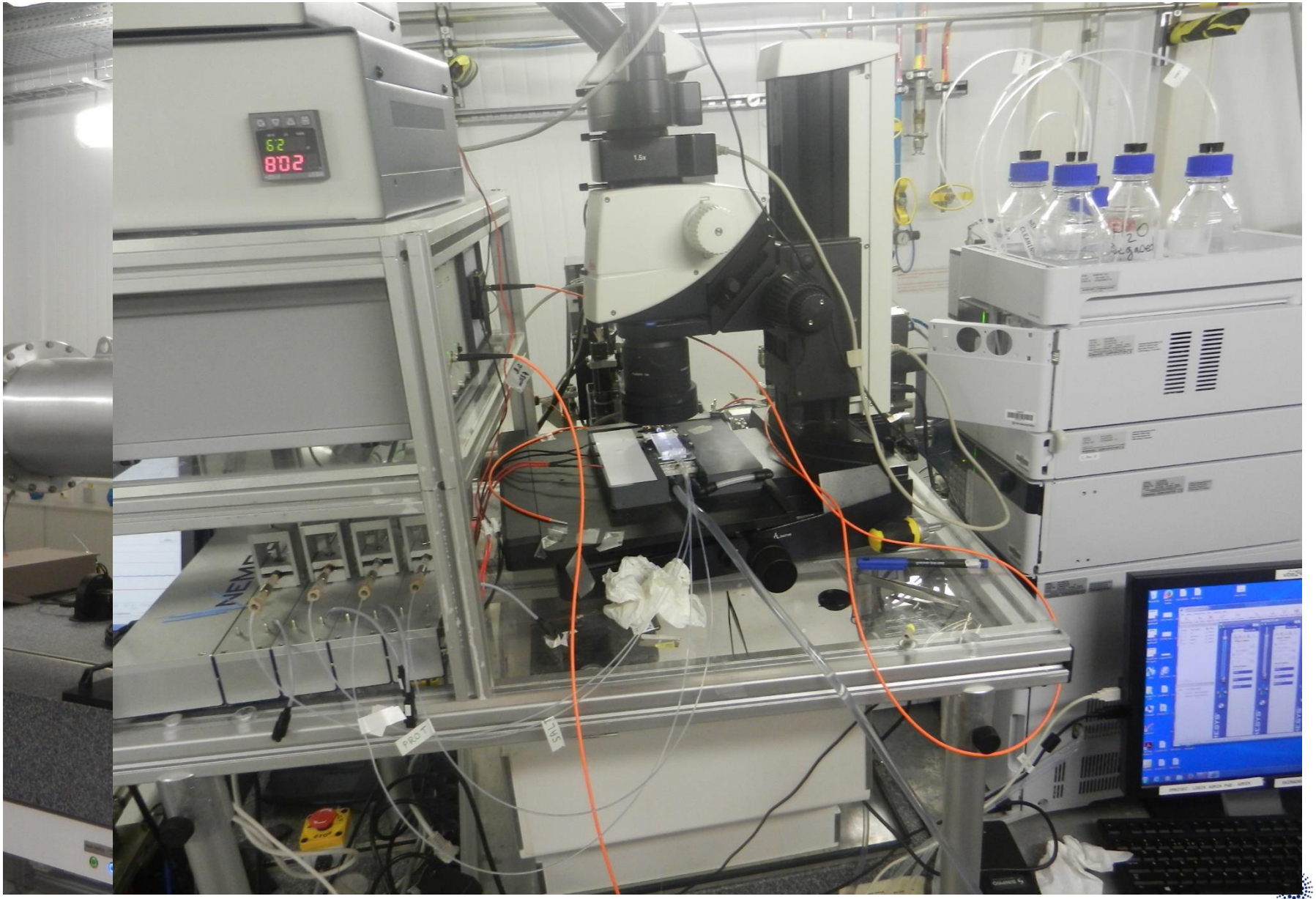
Samples stored in 96 well plates, PCR tubes
Thermo-regulation storage: 4-40ºC, exposure cell: 4-60ºC
Sample loading and cleaning: 30’



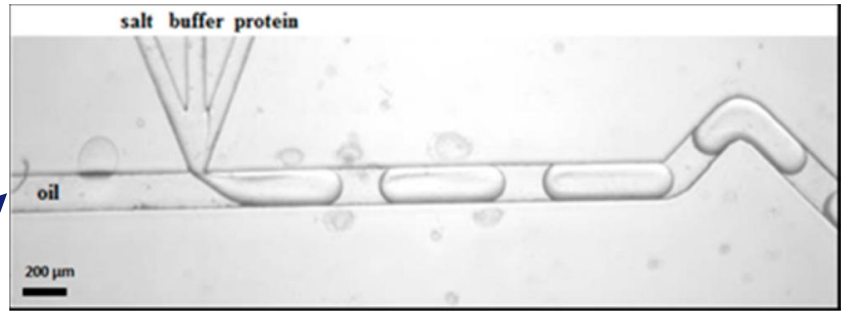
Switch between
SC and SEC modes
quick and robust

HPLC system mostly used in isocratic elution mode
(mobile phase composition is kept constant and uniform

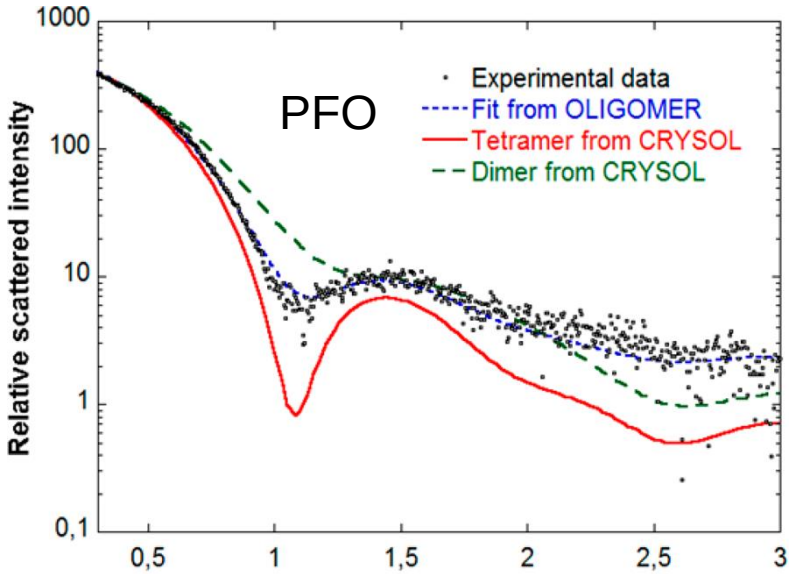
COOPLING SAXS AND PHOTONIC LAB-ON-A-CHIP SENSORS



MICROFLUIDICS DROPLET GENERATION – CHOICE OF SURFACTANT



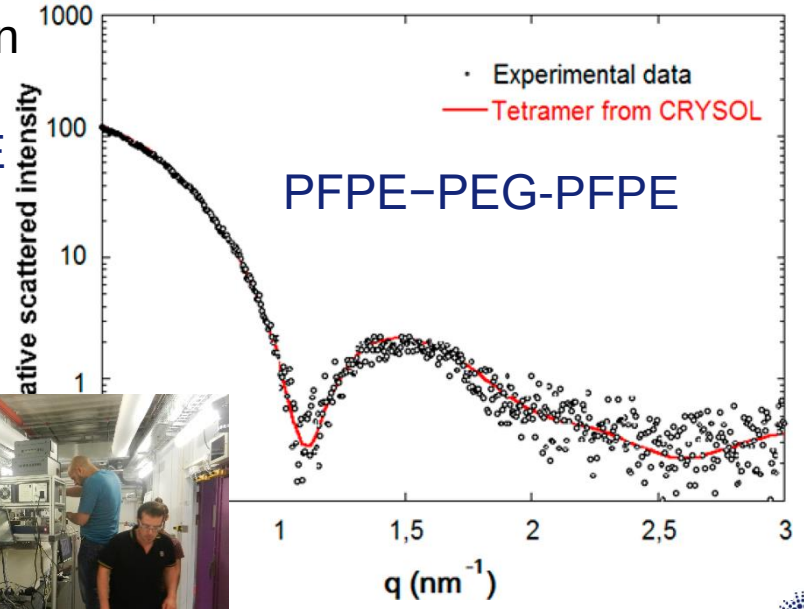
Krytox GPL100 oil + 2 wt % surfactant
(no radiation damage observed)



rasburicase

- high sensitivity to denaturation/dissociation
- with PFO (perfluorooctanol) surfactant
- with triblock copolymer PFPE−PEG-PFPE

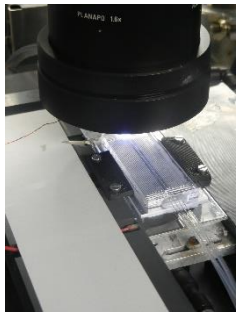
acts as a protectant of protein adsorption at the oil−water interface and avoids denaturation



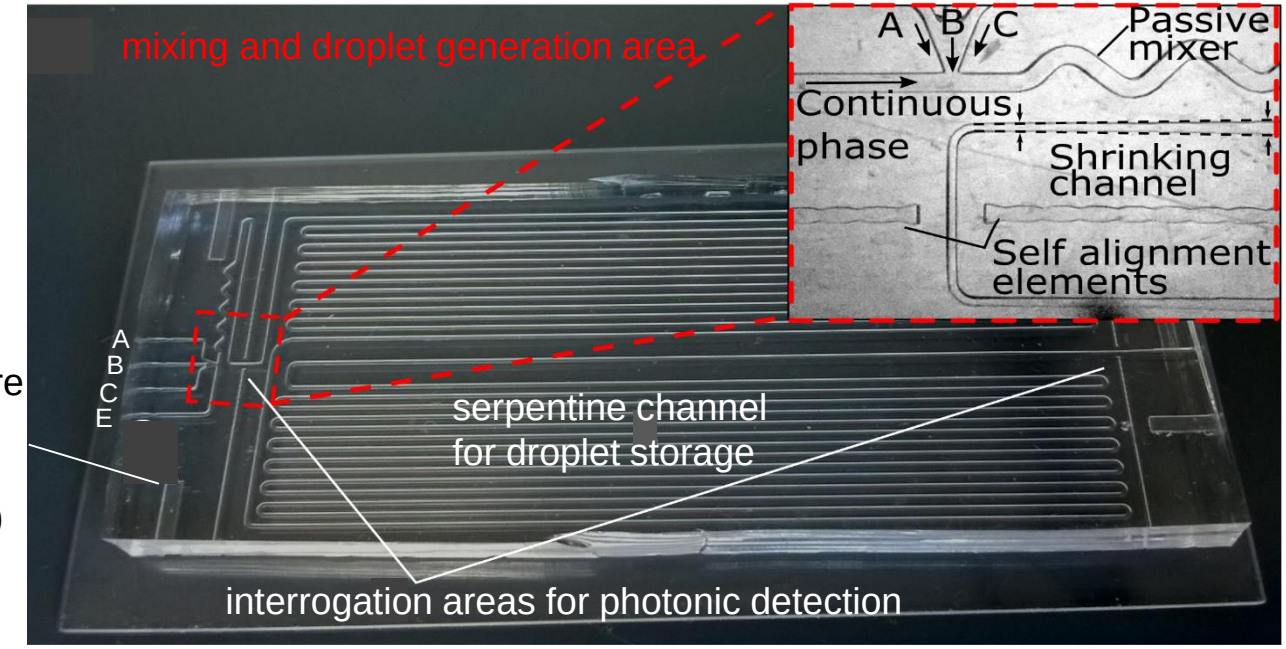
Nhat Pham, Dimitri Radajewski, Adam Round, Martha Brennich, Petra Pernot, Béatrice Biscans, Françoise Bonneté and Sébastien Teychené
Anal. Chem. ***89*** *(2017) 2282−2287*

COOPLING SAXS AND PHOTONIC LAB-ON-A-CHIP SENSORS

Protein solution droplets at different concentrations are generated and monitored in the PhLoC platform (rectangular channels of cross section of 280 x 300 μm^2)



inlet for temperature probes (to tune droplets reaction times from 0-50°C)



for pig-tailed fiber optics positioning

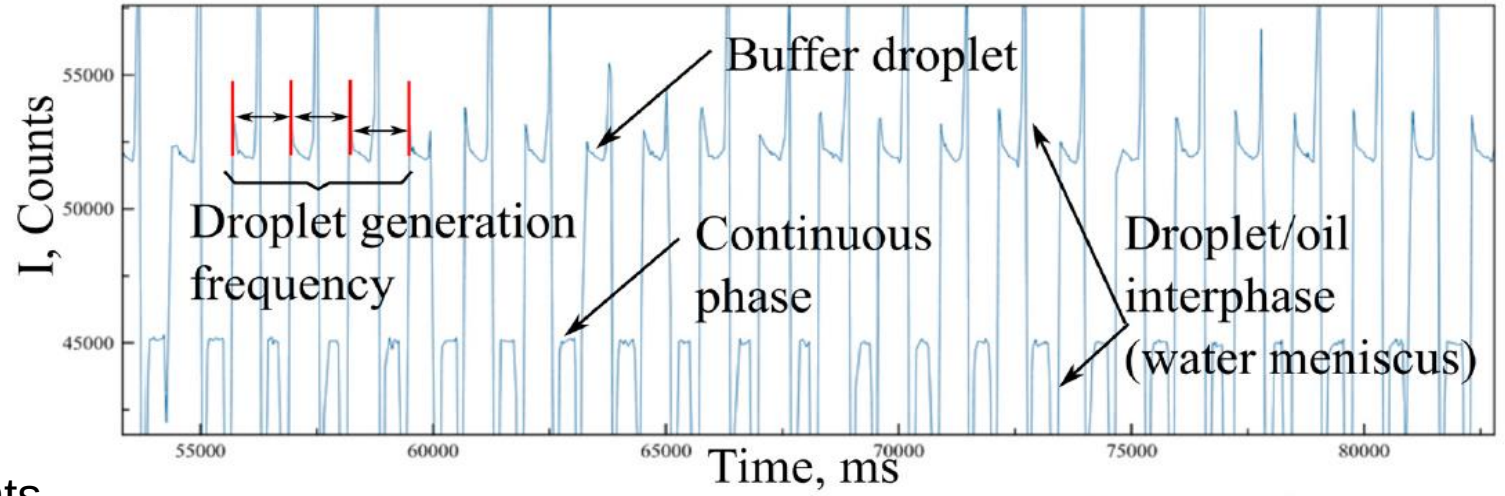
Isaac Rodríguez-Ruiz, Dimitri Radajewski, Sophie Charton, Nhat Phamvan, Martha Brennich, Petra Pernot, Françoise Bonneté and Sébastien Teychené
*Sensors **17** (2017) 1266-1278.*

Droplet generation and mixing:
- 3 channels for reagent injection (A, B, C),
- extra channel (E) for continuous phase injection,
- passive zigzag mixer allowing effective and fast droplet homogenization.

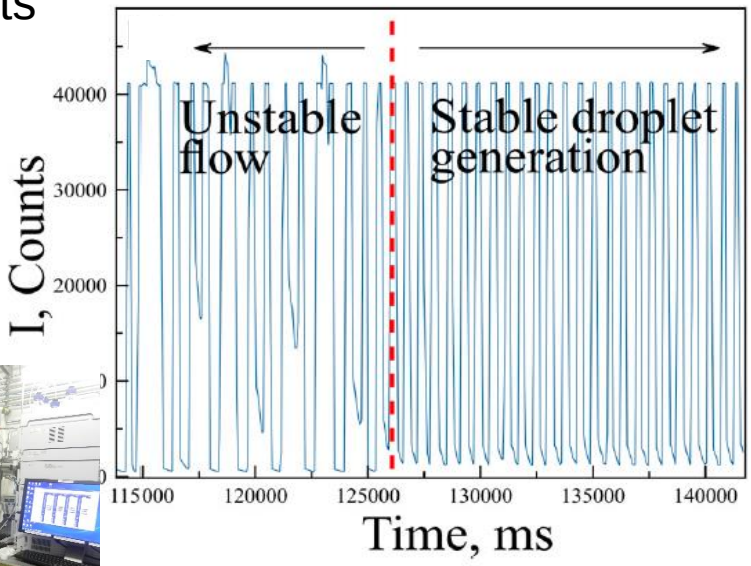
Link PhLoC-SAXS sample holder (300 μm Φ quartz) by a flexible fused silica capillary

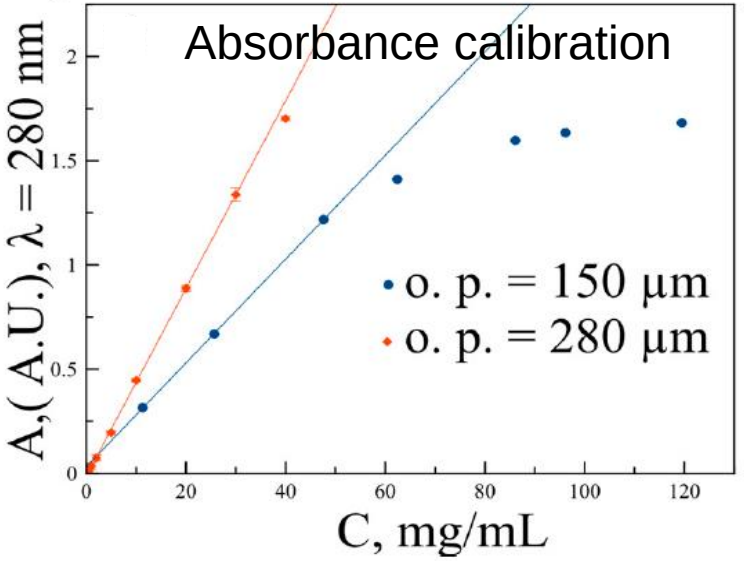
COOPLING SAXS AND PHOTONIC LAB-ON-A-CHIP SENSORS

Light intensity spectra at λ = 280nm when generating stable droplets at const. flow rate



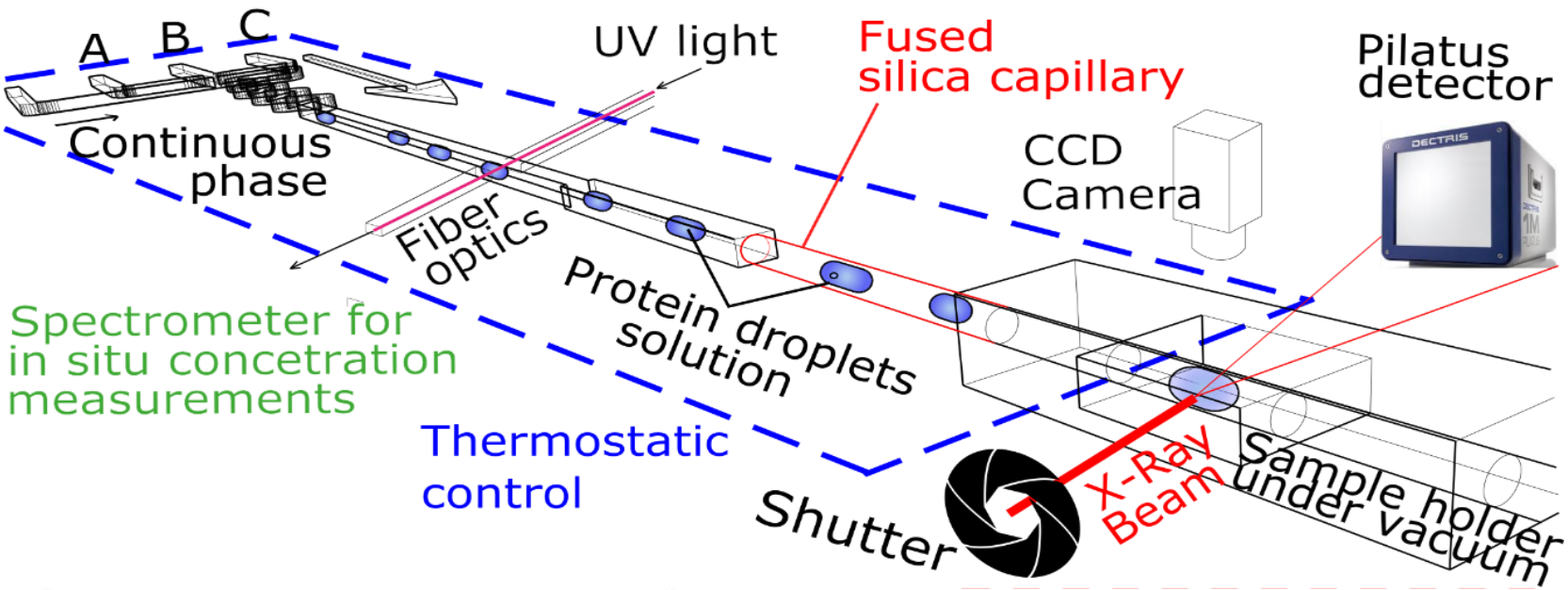
First instants of droplet generation

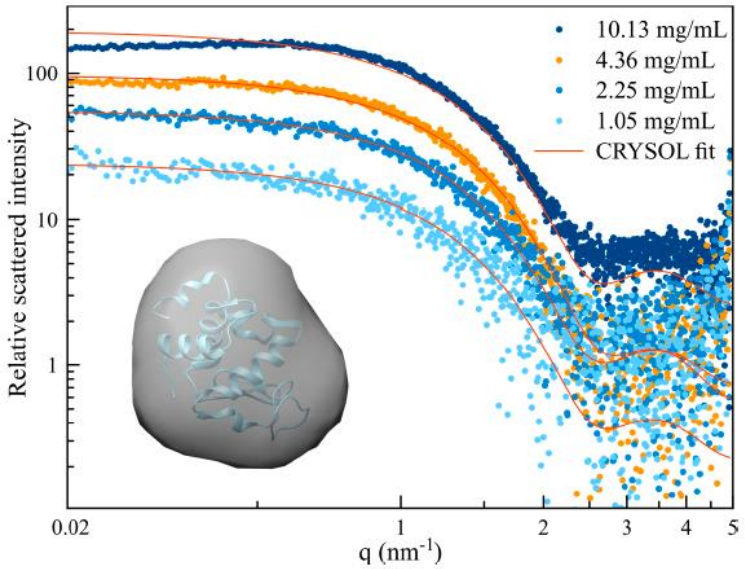




Lysozyme measured on chip through different optical path

COOPLING SAXS AND PHOTONIC LAB-ON-A-CHIP SENSORS





Normalized scattered intensity of lysozyme in NaAc 50 mM buffer pH 4.5 at different concentrations (average of 100 nano-droplets of same concentration).

Experimental and calculated scattering Intensity using CRYSOL for different protein concentrations, low resolution structure from orange curve (c = 4.36 mg/mL)

3D PRINTING FOR TESTING NEW SAMPLE HOLDERS

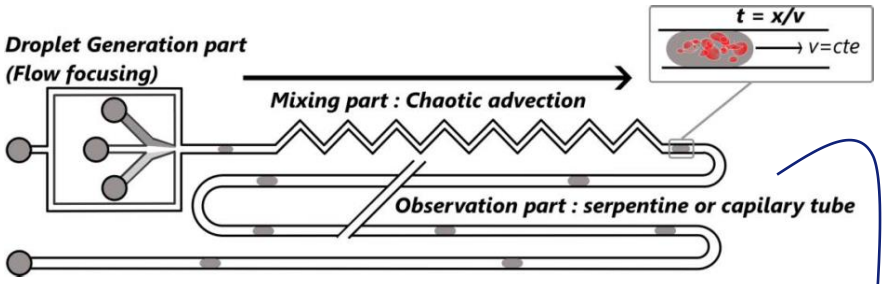
- vitreous carbon window, also mica or microfluidics chip

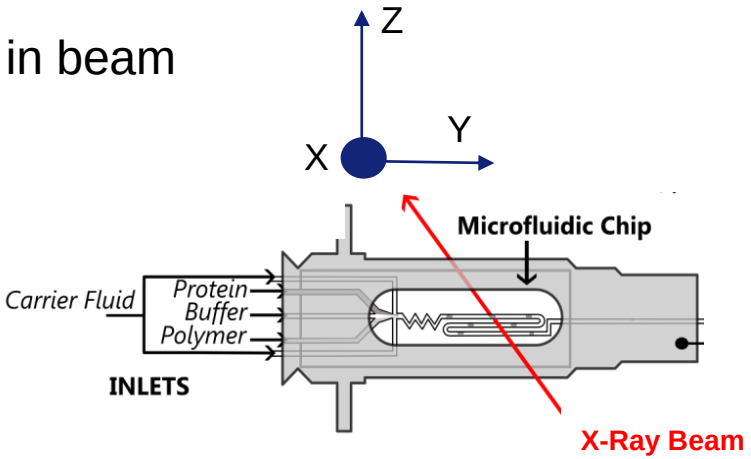
ASIGA Pico2 HD

Peter van den Linden
ESRF

MICROFLUIDICS WITH CHIP DIRECTLY IN X-RAY BEAM

In vacuum: chip is also sample holder, scan Y/Z in beam



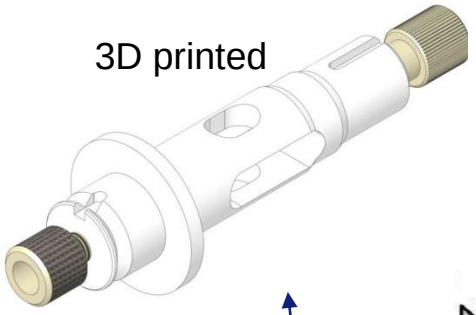


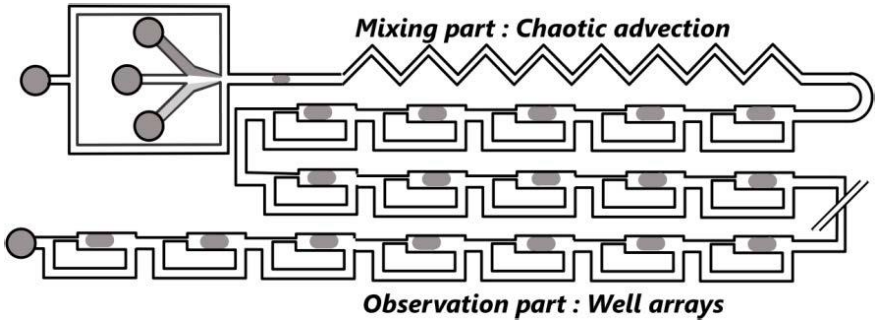
Fast nucleation *and growth:*
- distance in "serpentine" ~ time
- time scale down to 10 ms

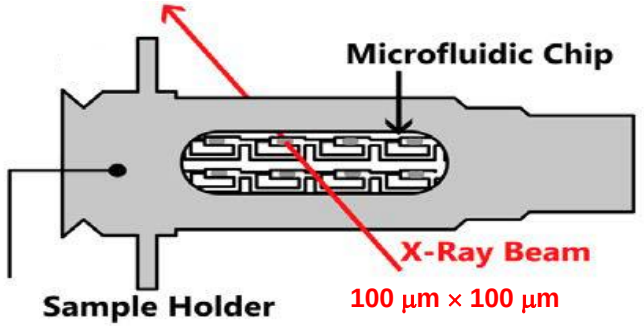
OR

Slow nucleation*:*
- droplets trapped in wells arrays
- measurement in each well to observe nucleation event

3D printed

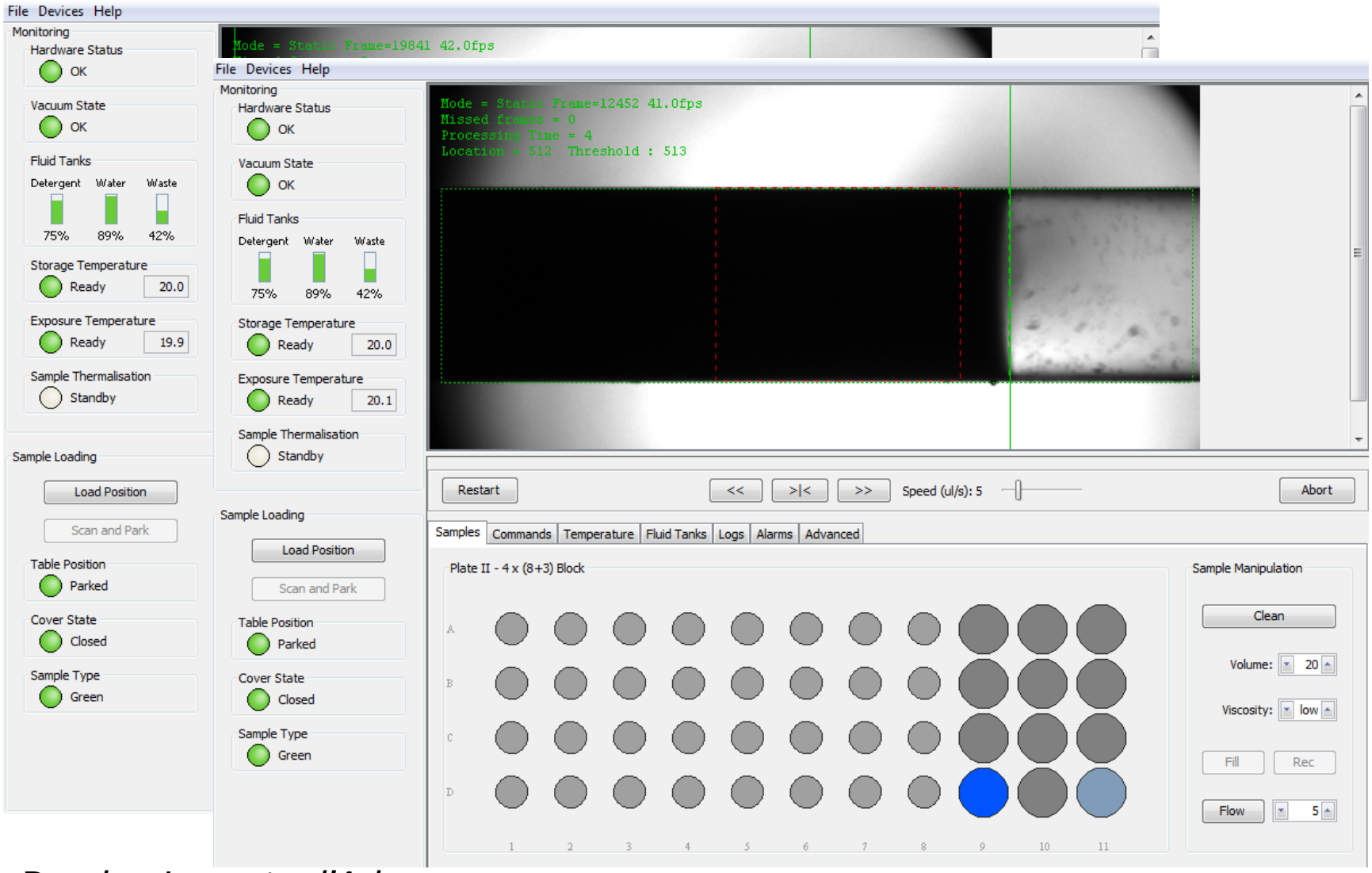






NON-TRANSPARENT SAMPLE DETECTION IMPLEMENTED

Dark is now detected…



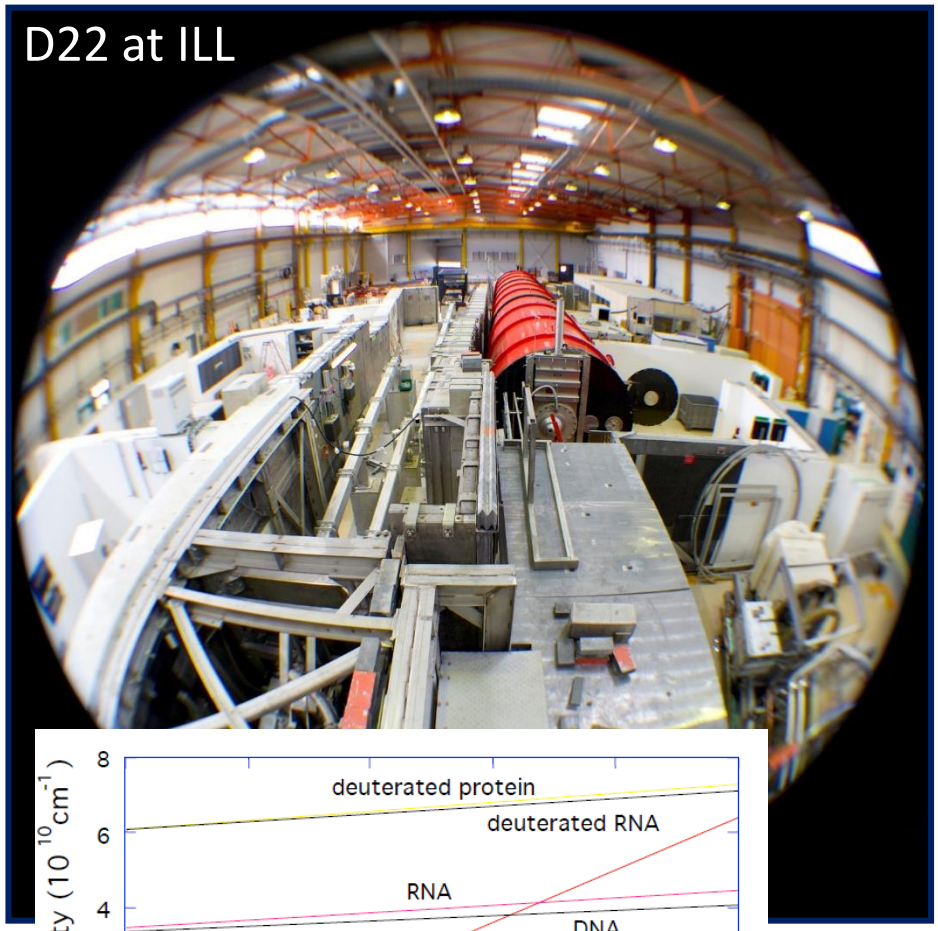
Damien Lacoste d'Arinax

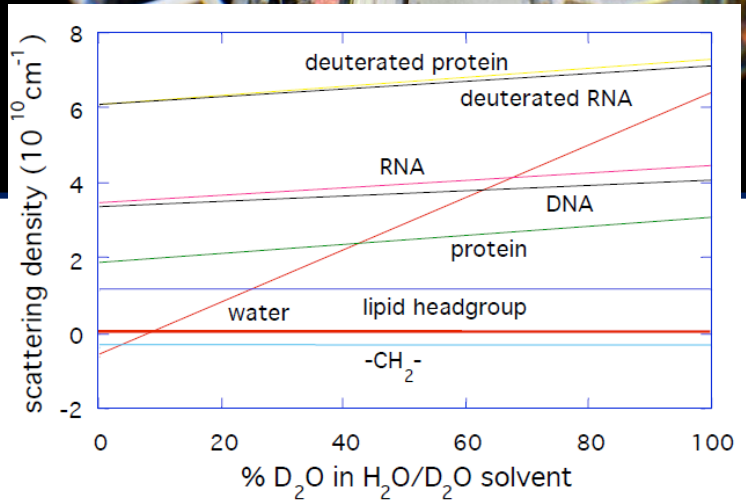
NEW ITEMS IN THE LAB

- HPLC pump: second SEC set-up in sample prep lab to equilibrate a column when running SAXS-SEC experiment in experimental hutch with other one

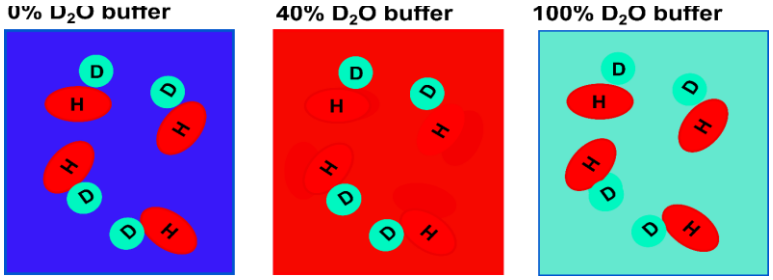
magnetic stirrer
miniSpin centrifuge

JOINT SANSAXS EXPERIMENT





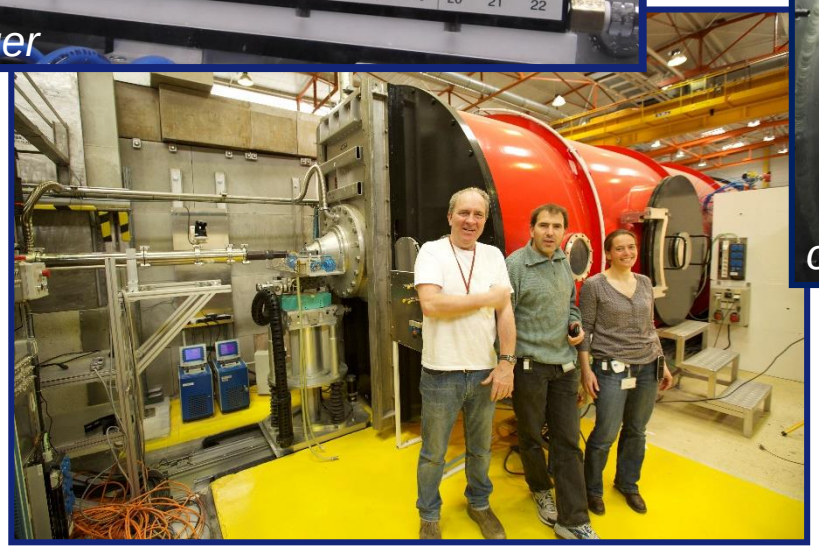
SANS contrast variation experiments obtained by exchanging the solvent for deuterated or partially deuterated solvent enhances the signal from one component of a complex.

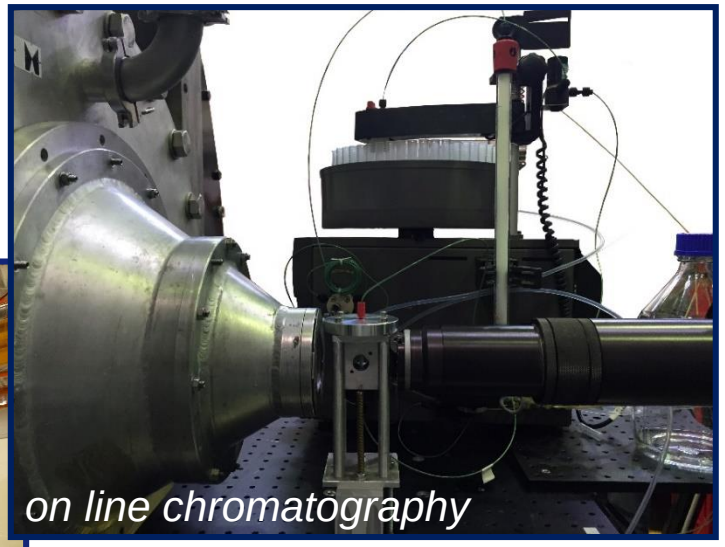


	SAXS	SANS
volume	small < 50 µl	larger ~ 300 µl
concentration	> 0.1 mg/ml	> 1 mg/ml
measuring time	short ~ s	longer ~ m÷h
radiation damage	yes	no
contrast variation	no	yes
sensitive to salts, denaturants	yes	no

Joint access – ESRF and ILL SAS experiments during one trip to Grenoble.

JOINT SANSAXS EXPERIMENT





SANSAXS BAG

from June 2016: **MX-1827** Trevor Forsyth, 9 shifts per period

- when SAN/XS proposal accepted and scheduled by ILL,
- beamtime, typically **1** shift, on BM29 within a few days of ILL experiment

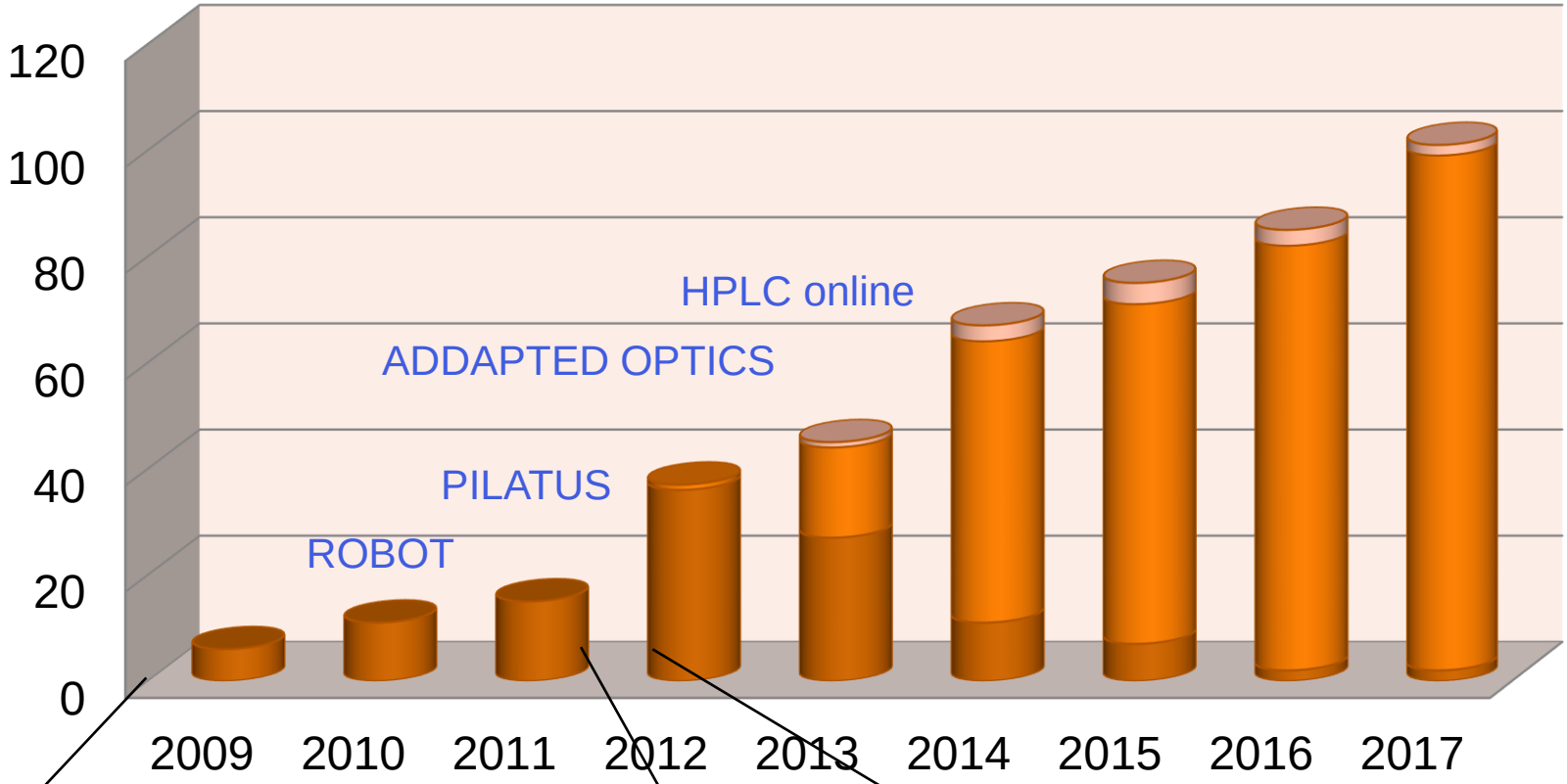
from Feb 2018: **MX-1987** Trevor Forsyth, 9 shifts per period

Actually performed Sansaxs experiments: 6 in 2016
3 in 2017 (long ILL shutdown Mar-Sept)

ESRF BIOSAXS PUBLICATIONS = RISE OF BIOSAXS

As on January 2018

$\Sigma_{TOTAL} = 442$



$\Sigma_{ID14-3} = 117$

$\Sigma_{BM29} = 324$

$\Sigma_{both} = 13$

First BioSAXS users at ID14-3 in November 2008

ID14-3 closed in December 2011

First BioSAXS users at BM29 in June 2012

MAIL _IN ROLLING PLANS/IDEAS

Motivations:

- needs to collect a few complementary data in very short time scale;
- beamline oversubscribed (nearly no place for Rollings);
- efficient use of available beam - in average per weak of beamtime:
  - ➡ 1 experiment cancelled by users (BAG exclusively);
  - ➡ 1 session of 3 shifts not using beam (no data collected) after late afternoon without any ‘good’ reason;
- less travelling;
- …

MAIL_IN Rolling

- ½ shift per month of beamtime scheduled (~ 3 shifts per period);
- no SEC-SAXS (needs appropriate column, equilibration);
- Sample Changer mode only with maximum 20 samples;
- safety sheets provided, experimental details, samples shipped and declared in ISPyB by user;
- LC can prepare dilution series if samples can not be shipped already in stripes or plates;
- …