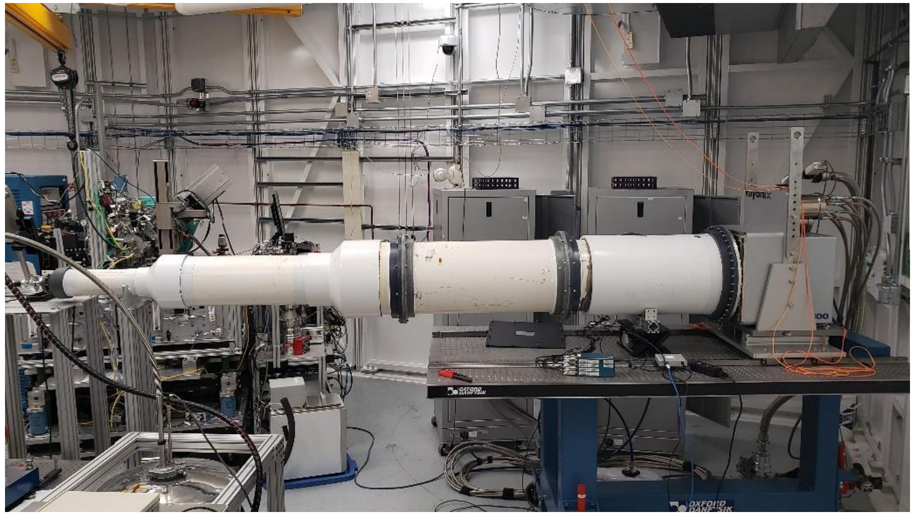
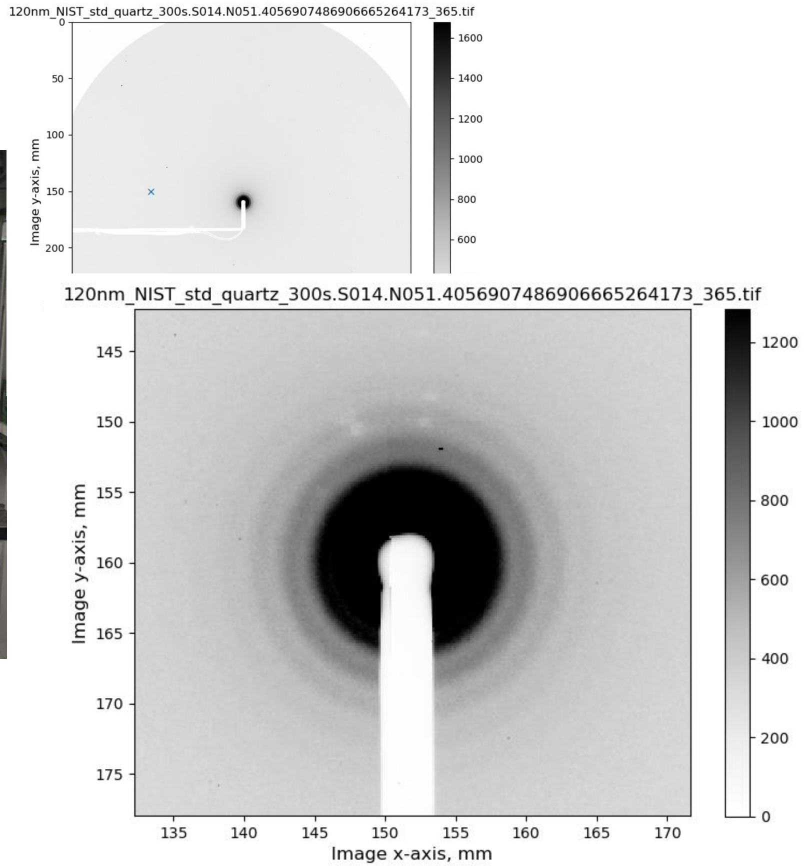
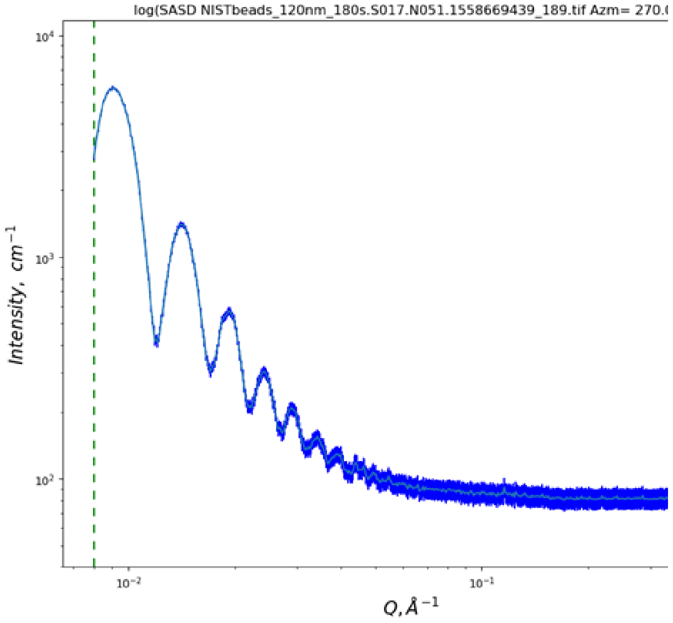
An Introduction to Small Angle X-ray Scattering (SAXS)







Adam Leontowich
CLS XRD school, Thursday, June 20, 2024

Canadian Light Source
Centre canadien de rayonnement synchrotron

The small angle X-ray scattering (SAXS) region

Instrumentation, sample prep, data collection and reduction

Basic data processing with examples from BXDS

Brockhouse X-ray Diffraction and Scattering (BXDS) sector

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The lower energy diffraction and scattering side-bounce beamline for materials science at the Canadian Light Source

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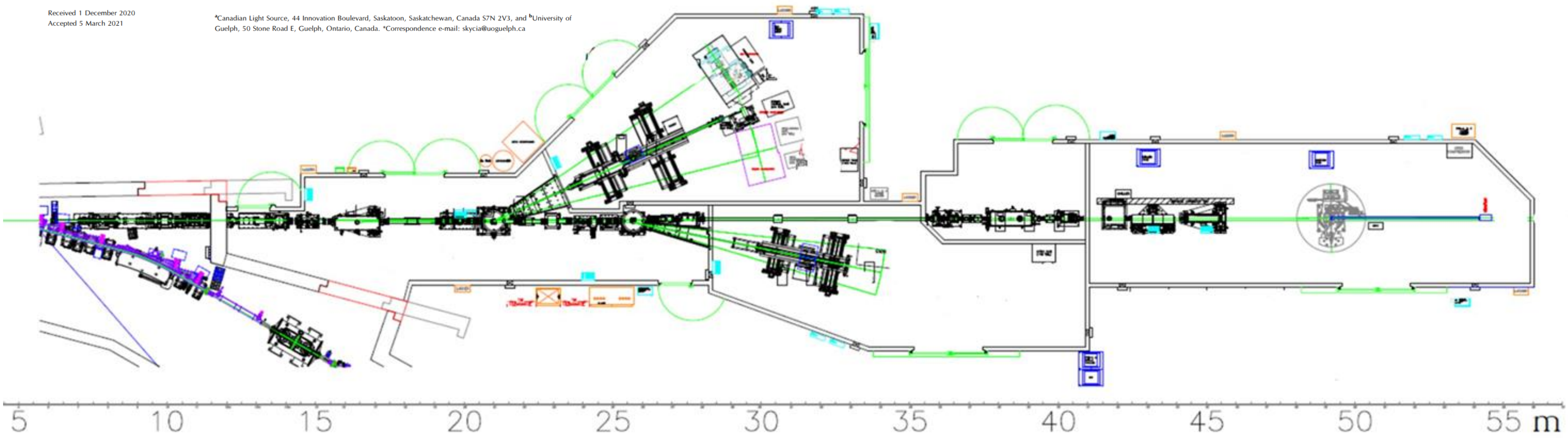
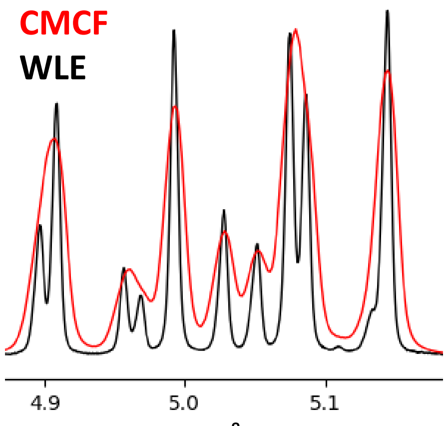
3 “hard X-ray” beamlines (5 – 100 keV)



WLE beamline

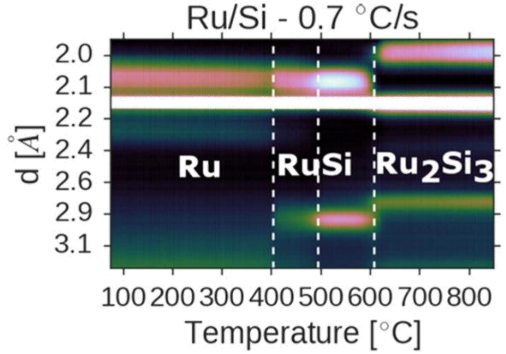
High-resolution powder diffraction



- Peak shape analysis
- Complex mixtures
- Complex structures

IBM (rapid thermal annealing to 1100 °C)

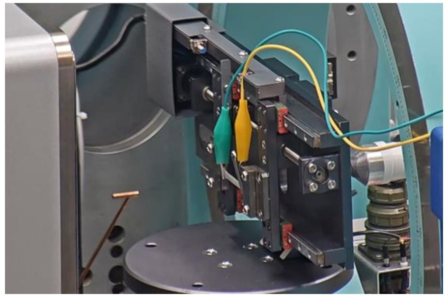
- Thin film studies: XRD, resistivity, roughness, under ultra-high purity gas



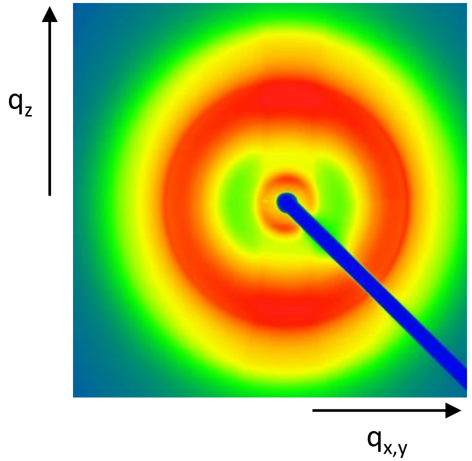
S. Dey et al., J. Vac. Sci. Technol. A 35, 03E109 (2017)

WAXS (XRD with area detector)

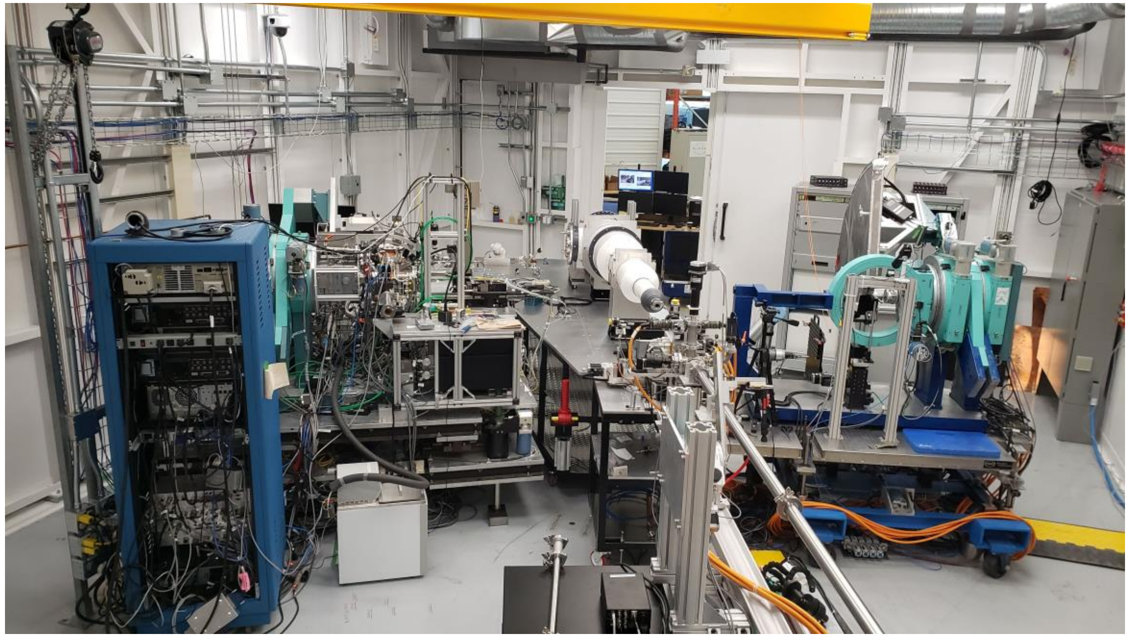
Biopolymers for bandages
Youchao Teng, Yimin Wu, U. Waterloo



Measure WAXS, voltage and current while stretching (0 – 100 N)



- Texture on surfaces
- Degree of orientation
- % crystallinity
- Speed/in-situ



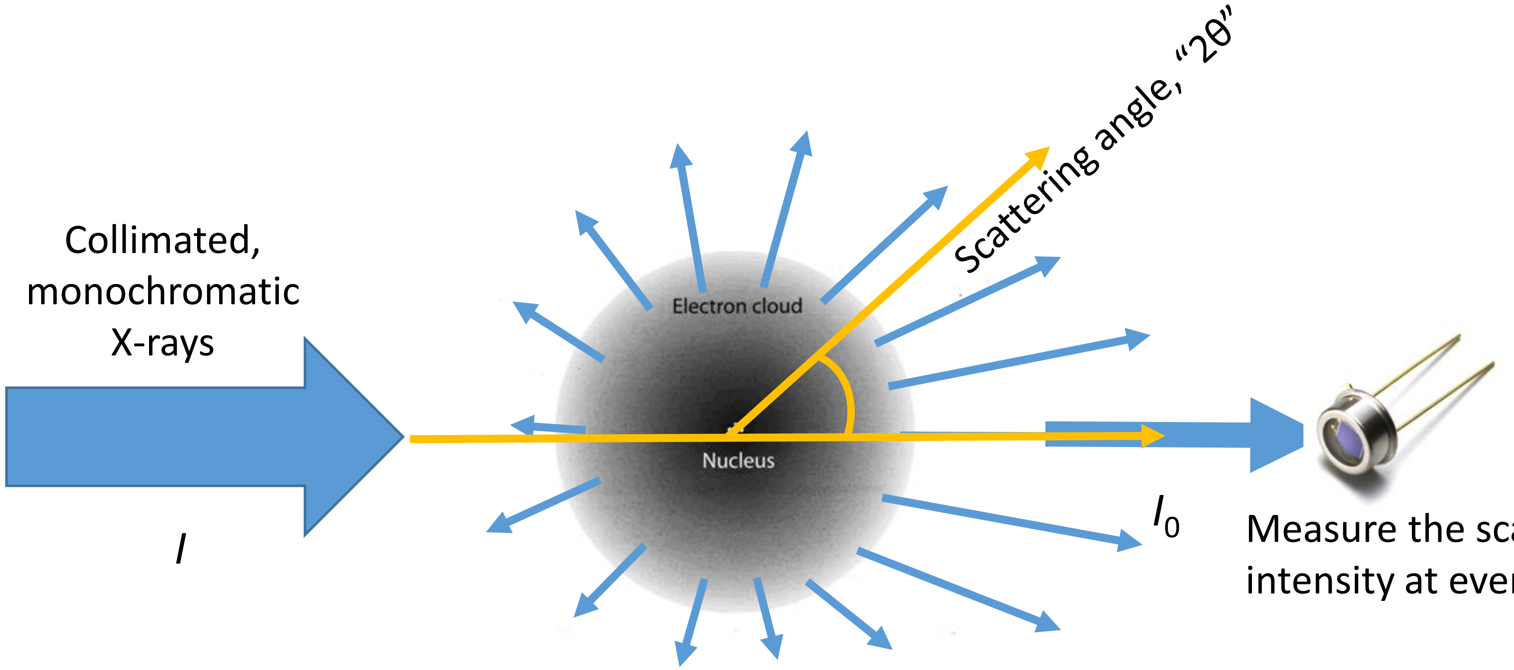
SAXS/WAXS

SAXS has been running since February 2021

No dedicated SAXS beamline at CLS

X-ray scattering

Collimated, monochromatic X-rays



Measure the scattered intensity at every 2θ

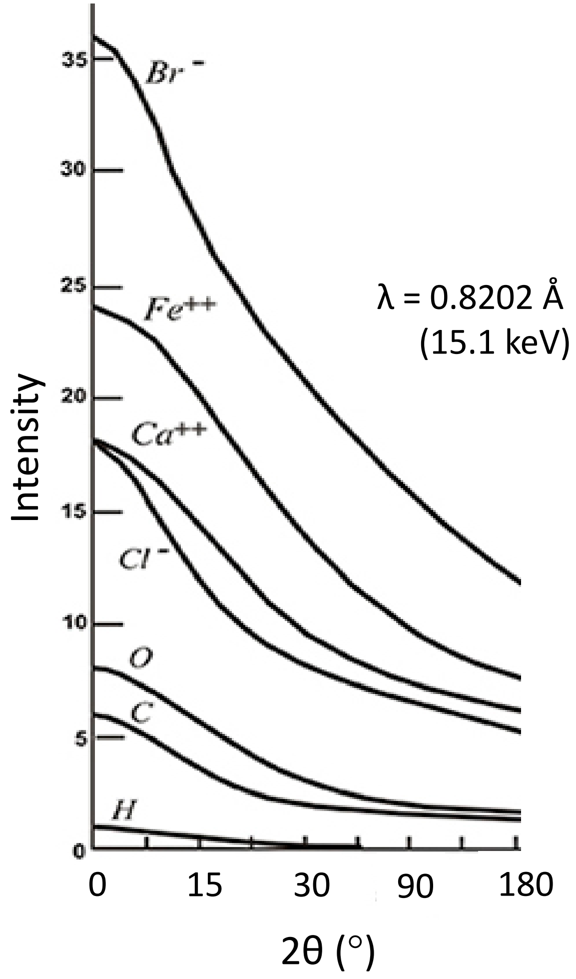
Photoabsorption

$T = I/I_0 = e^{-(\mu/\rho)\rho t}$

"Elastic" scattering

no wavelength change

Atomic form factors
(or atomic scattering factors)



From scattering to diffraction

"Bragg" diffraction

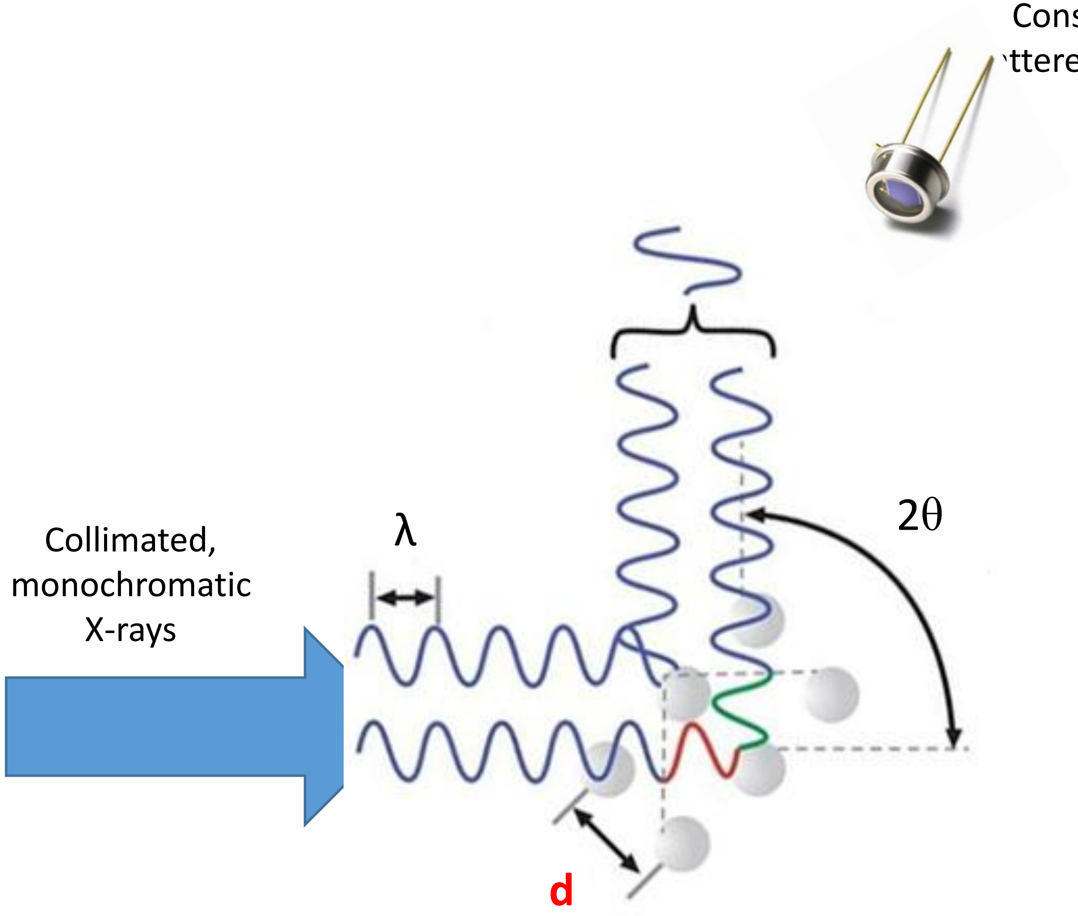
Constructive interference channels scattered intensity into specific directions

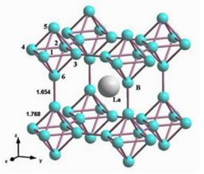
$n\lambda = 2\mathbf{d}\sin\theta$

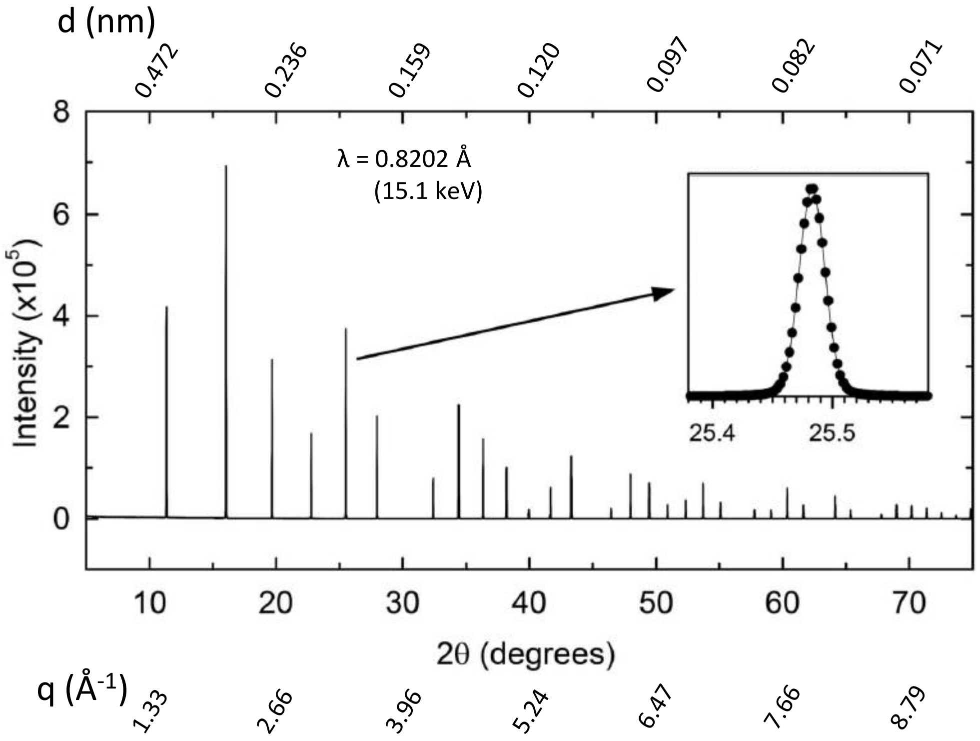
2θ ↔ **d** ↔ **q**

$q = (4\pi\sin\theta)/\lambda$

$q = 2\pi/d$





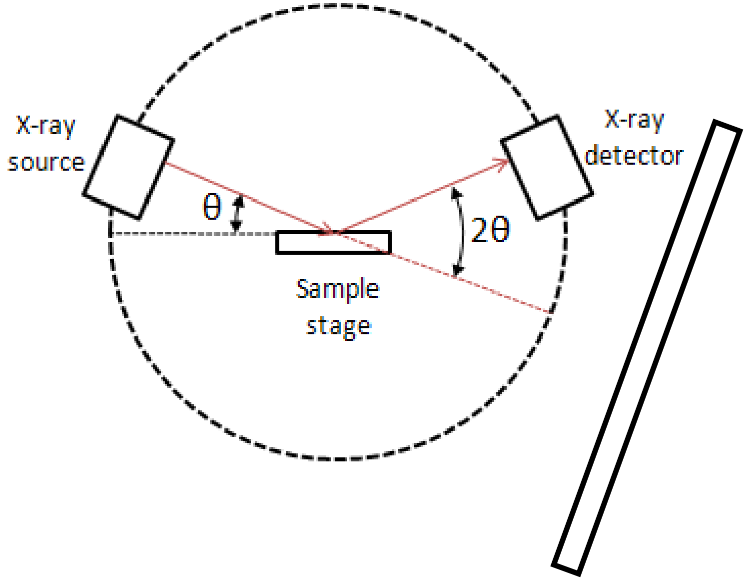


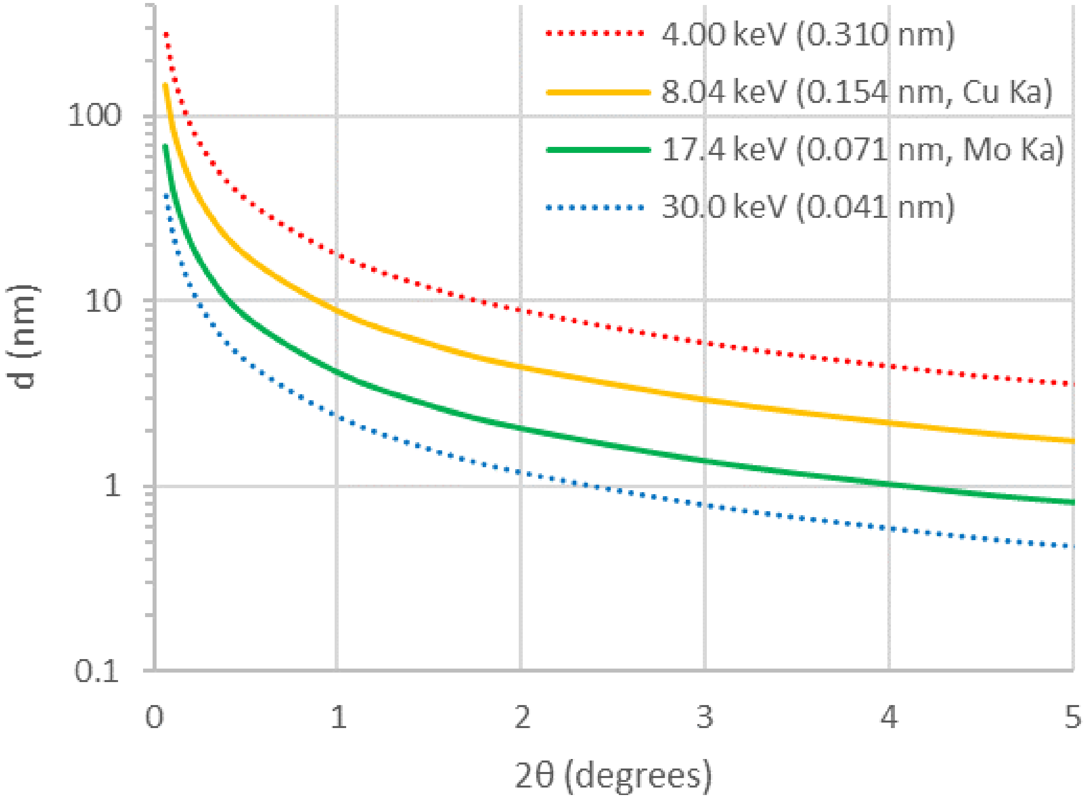
X-ray scattering and diffraction reveals structural order within materials on the atomic to 100s of nanometer length scale

What is SAXS all about?

$\lambda = 2d\sin\theta$

Starting from powder diffraction…





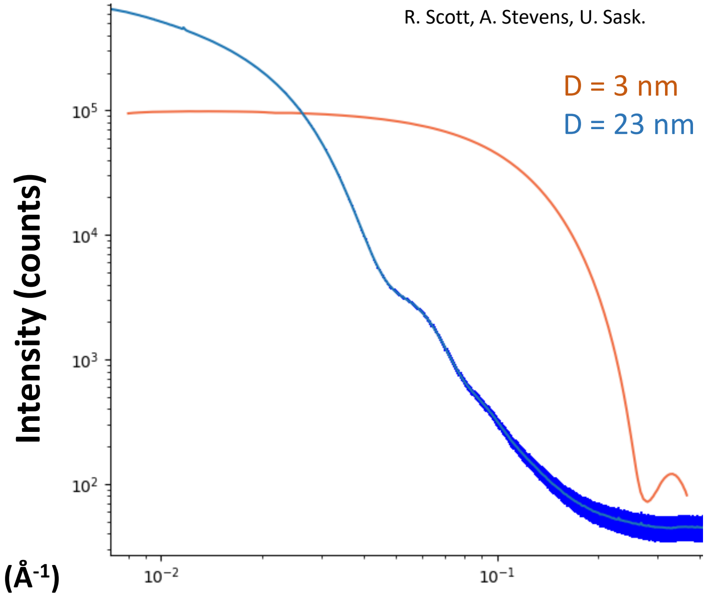
The nano world

Atomic distances, spacing between planes of atoms, bond lengths

SAXS is generally 2θ = 0.05 - 5°

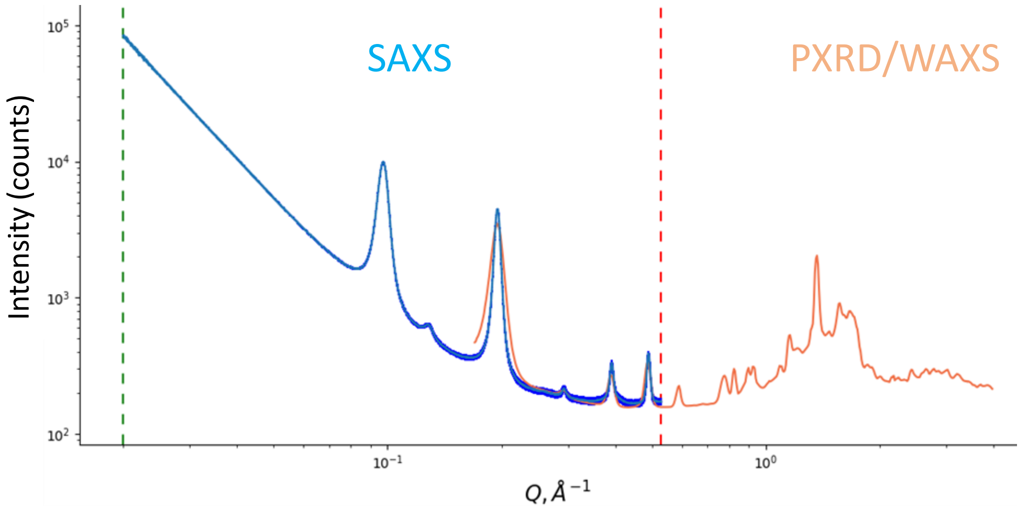
Probe relatively big things (1 - 150 nm) by measuring elastic X-ray scattering at small angles

How is SAXS related to other X-ray diffraction techniques?

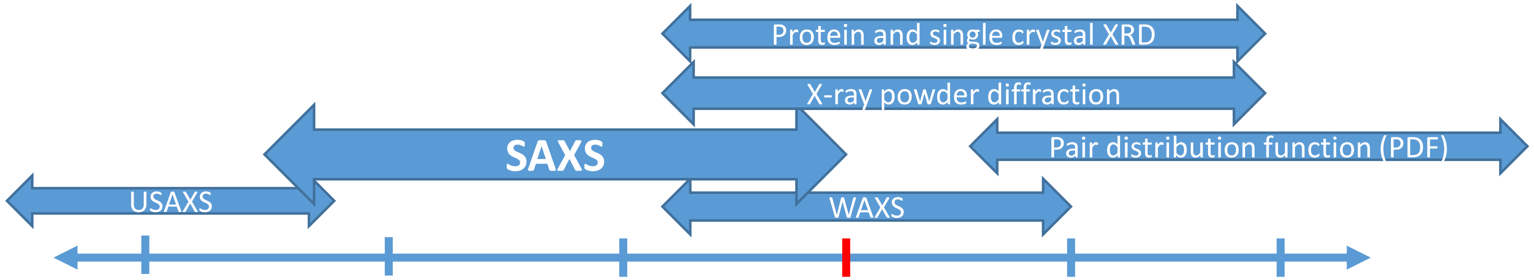


Q (Å⁻¹)	10^{-2}	10^{-1}
d (nm)	62.8	6.28
2θ (°)	0.108	1.08

- Intensity vs. Q
- log-log plot



Chocolate
A.G. Marangoni, U. Guelph



λ = 0.819 Å
(15.1 keV)

Q (Å⁻¹)	0.000603	0.00603	0.134	0.67	5.87	9.87
d (nm)	1042	104	4.69	0.938	0.106	0.0637
2θ (°)	0.0045	0.045	1	5	45	80

Nano-scale

Inter-molecular distances and packing arrangements, particle size and shape

Atomic-scale

Inter-atomic distances, bond lengths, nearest neighbor atoms
Crystal structures with atomic resolution

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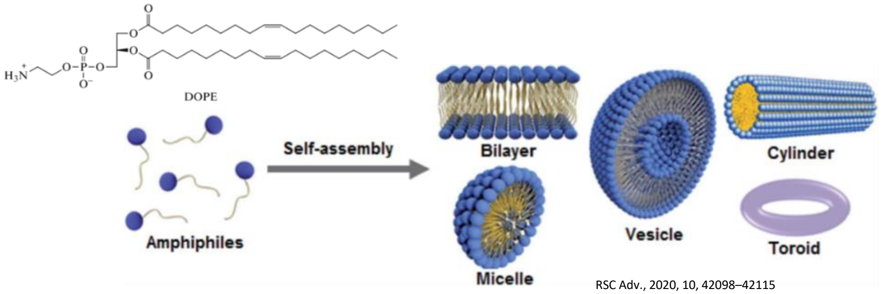
Applications

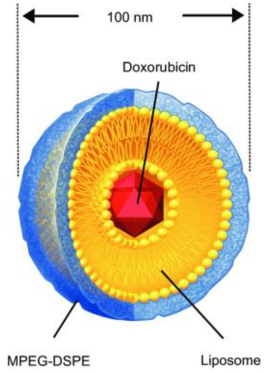
Amphiphiles



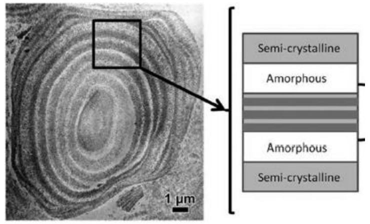
RSC Adv., 2020, 10, 42098–42115

Drug delivery,
Pharmaceuticals

Micelles,
emulsions,
surfactants…

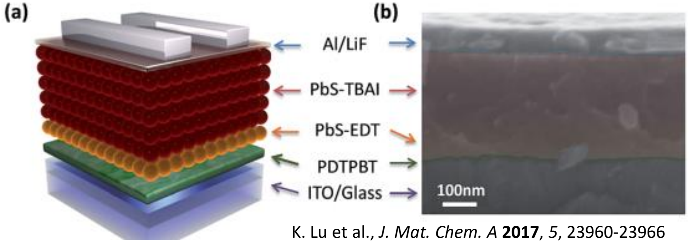


Food science

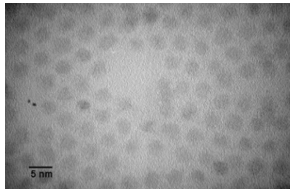


S. Perez and E. Bertoft, *Starch* **2010**, *62*, 389 - 420

All kinds of nanoparticles



K. Lu et al., *J. Mat. Chem. A* **2017**, *5*, 23960-23966

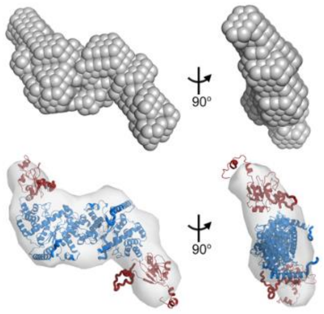


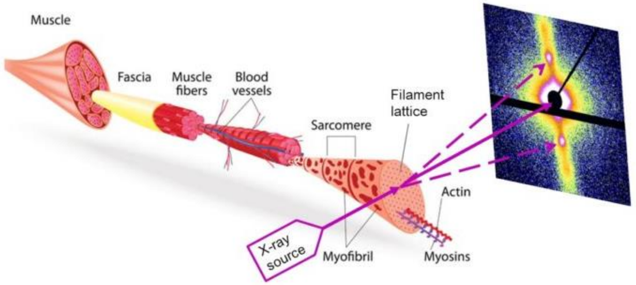
M. Yuan et al., Adv. Mater. 2014, 26, 3513–3519

Biomacromolecules

Cellulose
Collagen
Chitosan

Proteins





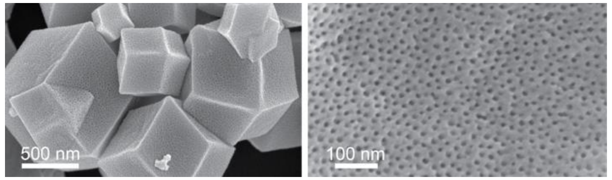
https://www.techexplorist.com/scientists-investigated-pork-fillet-x-ray-light/11572/

Nano-porous materials

Geology

Batteries

Zeolites



M. Liu, Z.M Hudson, UBC

1) Packing, ordering
2) Size, size distribution
3) Shape

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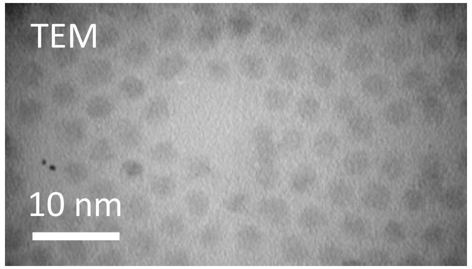
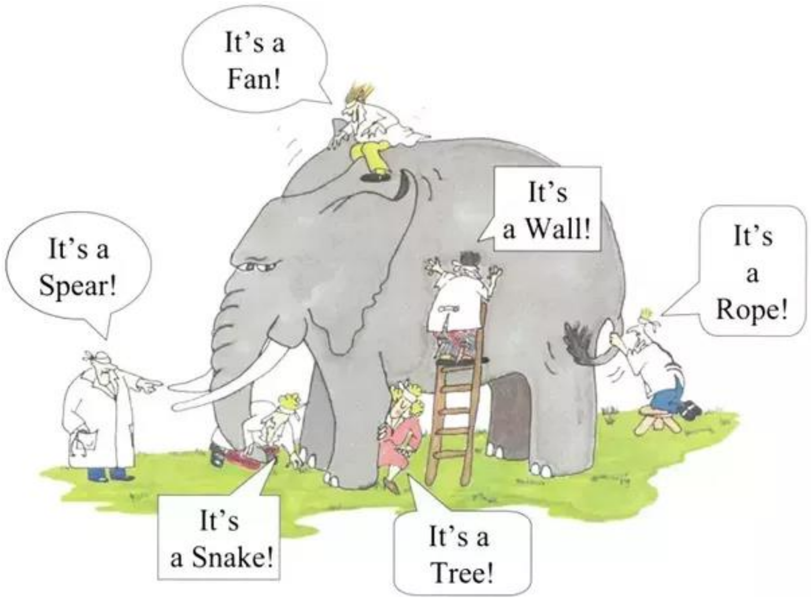
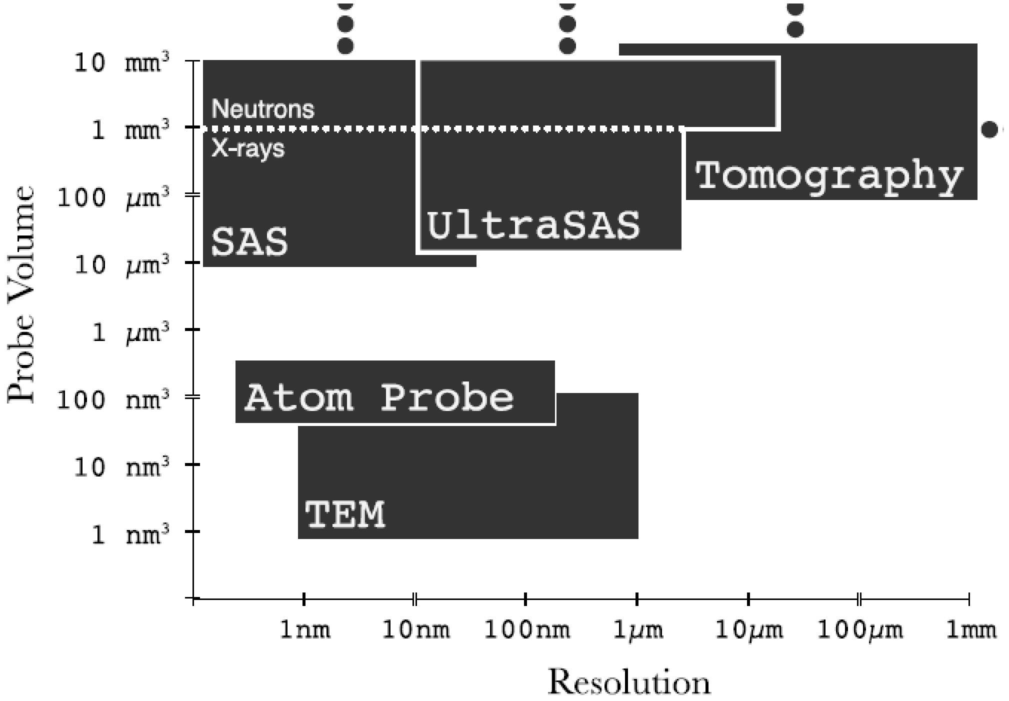
SAXS is highly complementary to microscopy



M. Yuan et al., Adv. Mater. 2014, 26, 3513–3519



"Blind men and an elephant"



B.R. Pauw, J. Phys.: Condens. Matter 25 (2013) 383201

- FOV decreases as resolution increases
- Observe a small fraction of the complete sample at a time
- Challenging sample prep at smaller size

➢ SAXS provides nanoscale information **averaged** over the beam volume (~mm^3)
➢ Complementary info: shape, folding/unfolding, assembled state in solution

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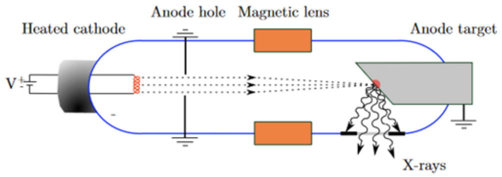
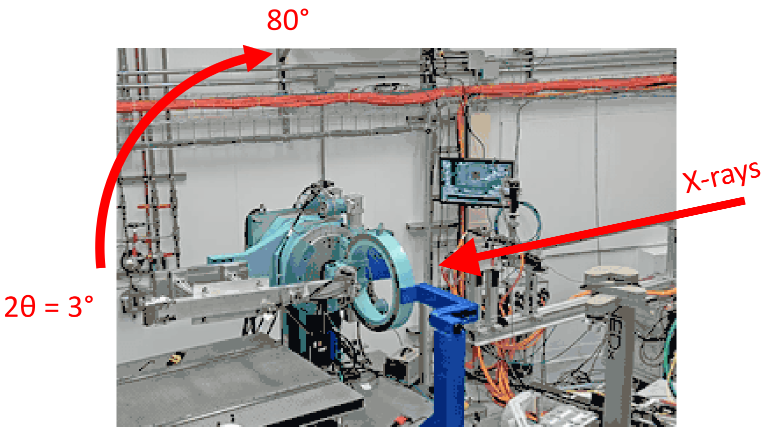
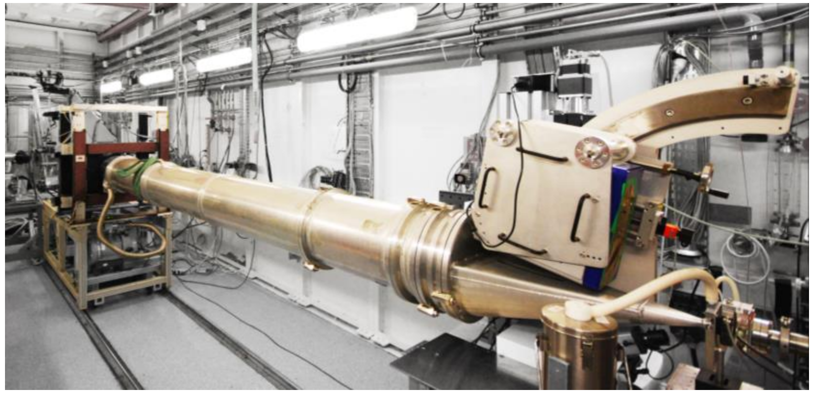
Instrumentation, sample prep, data collection and reduction

How to measure small angle X-ray scattering?

Lab source



Synchrotron

	PXRD	SAXS
Lab source		
Synchrotron		ESRF - BM26 

1) Special instrumentation required for SAXS
2) SAXS instruments are long!

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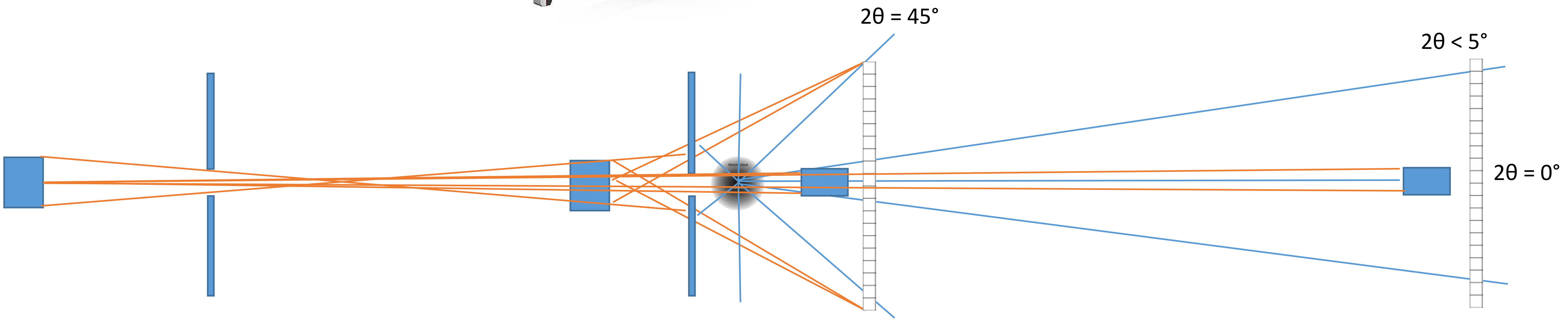
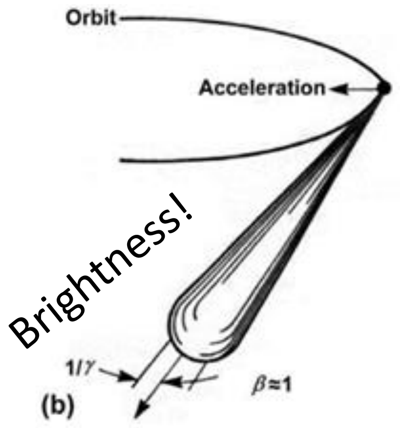
SAXS instrumentation basics





- More flux (better SNR), weakly scattering objects (organics/biology)
- Better q resolution
- Time resolved, SAXS mapping
- Choice of many wavelengths

- Highly collimated hard X-ray beam (multiple slits)
- Large area detector with beamstop
- Space for >2 m sample to detector distance
- Some ability to change detector distance (SAXS/WAXS)
- All or most components in vacuum

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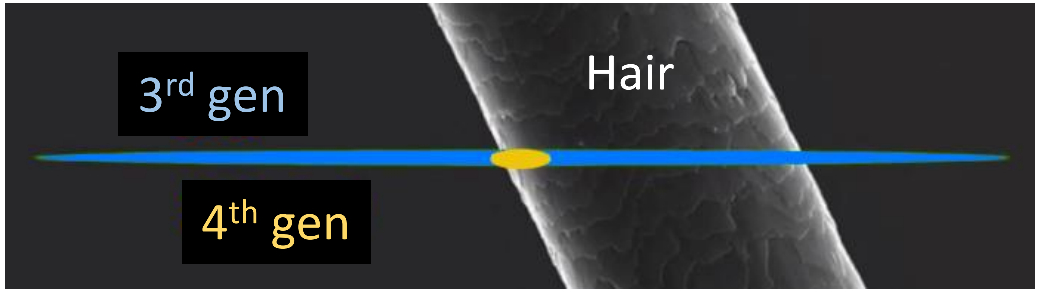
State of the art instrumentation

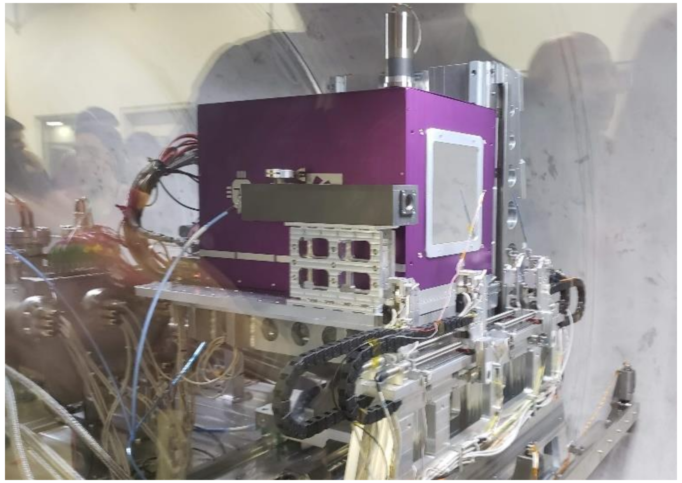


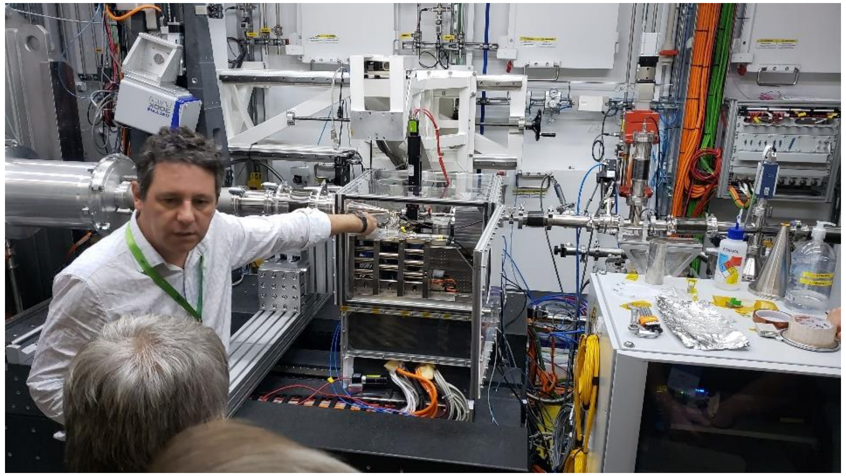
P. R. Willmott, 2022

CATERETÊ beamline @ SIRIUS
Campinas, Brazil

Dedicated SAXS beamline, new for 2022

- 4th gen synchrotron, 88 m source to sample
- Modern large area detector with beamstop
- 0 - 28 m sample to detector distance
- All components in vacuum, no windows from source to detector





SAXS user facilities in Canada

CLS

SFU

SAXSLAB, Ganesha 300XL+

https://www.4dlabs.ca/our-capabilities/equipment/small-angle-xray-scattering-system.html

McGill

Anton Paar, SAXSpoint 2.0

https://www.mcgill.ca/mc2/instrumentation/x-ray-diffraction/saxs

U. Montreal

SAXSLAB, Ganesha

http://airen.bcm.umontreal.ca/biostruct/scheduling_saxs/

U. Alberta

Bruker D8 DISCOVER Plus

https://www.nanofab.ualberta.ca/2023/news/saxs-gi-saxs-waxs-are-available-on-bruker-d8d-plus/

U. Toronto

Anton Paar, SAXSpace

https://lab.research.sickkids.ca/sbc-facility/saxs/

McMaster

MacSANS - Commissioning begins mid-July 2024

MacSANS Overview

Source-to-Sample	14 m
Sample-to-Detector	2 – 10 m continuous
Peak Thermal Flux on Sample	10^4 - 10^5 neutrons/cm^2/s
Neutron Wavelength	2.6 - 23 Å at Δλ/λ ≈ 17%
Detector	100 cm^2 at 0.9 cm
Accessible Q-range	0.005 < Q < 1.25 Å^{-1}
Structure Scale	0.6 – 125 nm

https://nuclear.mcmaster.ca/neutron-beams/

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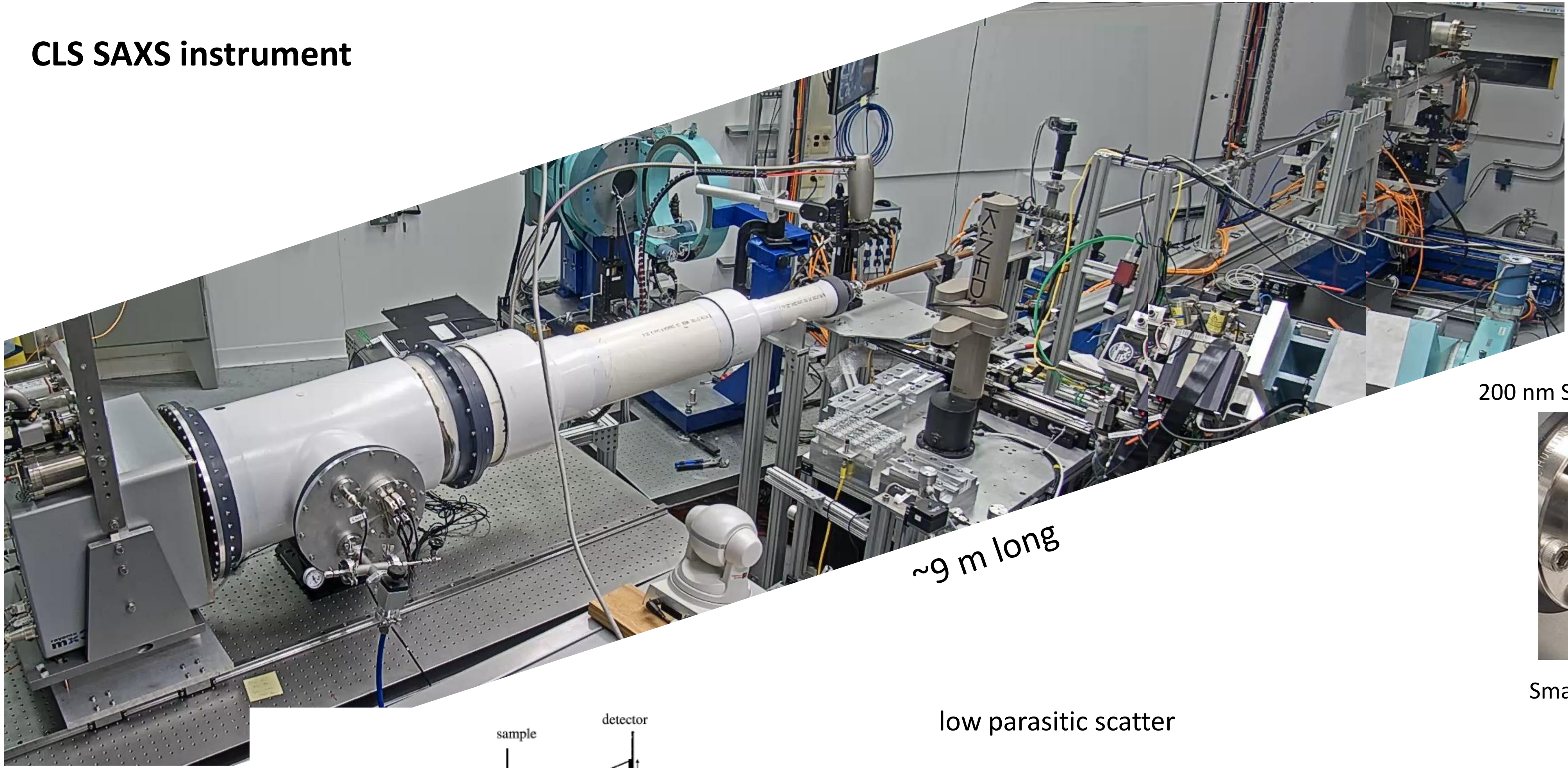
CLS SAXS instrument

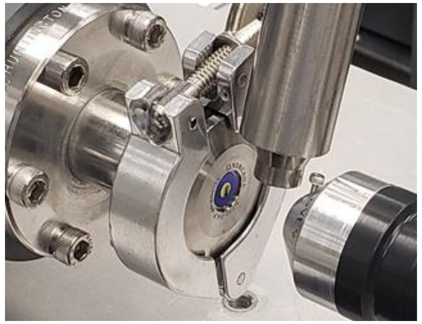


1) Beam defining slit
+6.1 m
(1.00 mm x 1.00 mm)

2) Anti-scatter slit
+0.9 m
(0.25 mm V x 0.50 mm H)

200 nm SiN window

3) Guard slit (0.80 mm)



Small air gap for sample (~35 mm)

3 pinhole layout

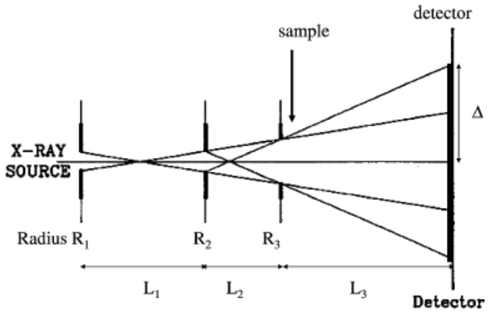
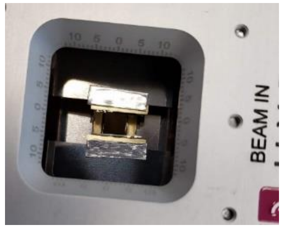
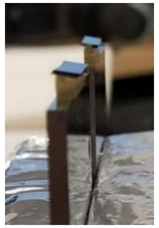


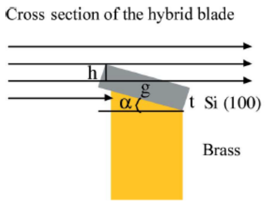
Figure 4
Schematic layout of a SAXS pinhole camera.

J. Appl. Cryst. (2004). 37, 369-380

low parasitic scatter

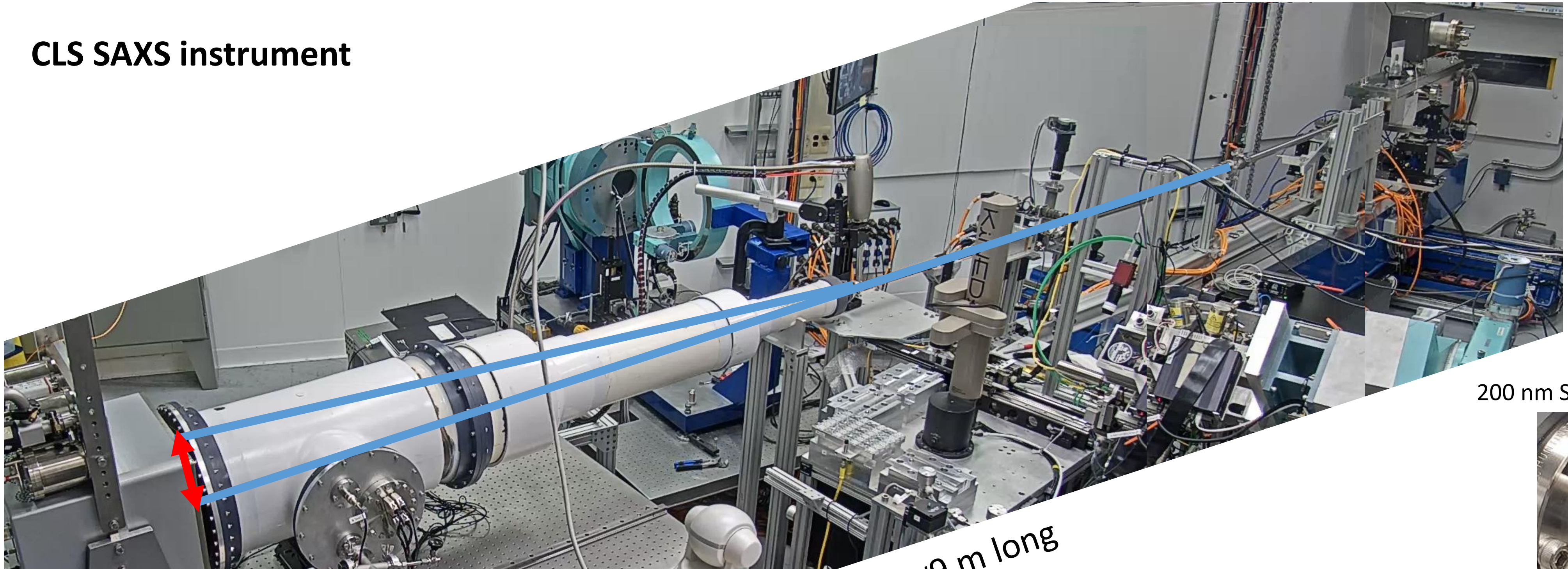






Y. Li et al., J. Appl. Cryst. (2008). 41, 1134–1139
N.M. Kirby et al., J. Appl. Cryst. 46, 1670–1680 (2013)

CLS SAXS instrument



~9 m long

λ = 1.26 Å (9.85 keV)
0.658 Å (18.8 keV)

Q ~ 0.005 – 0.5 $Å^{-1}$
d ~ 125 – 1.25 nm
2θ ~ 0.05 – 3.7° (see website)

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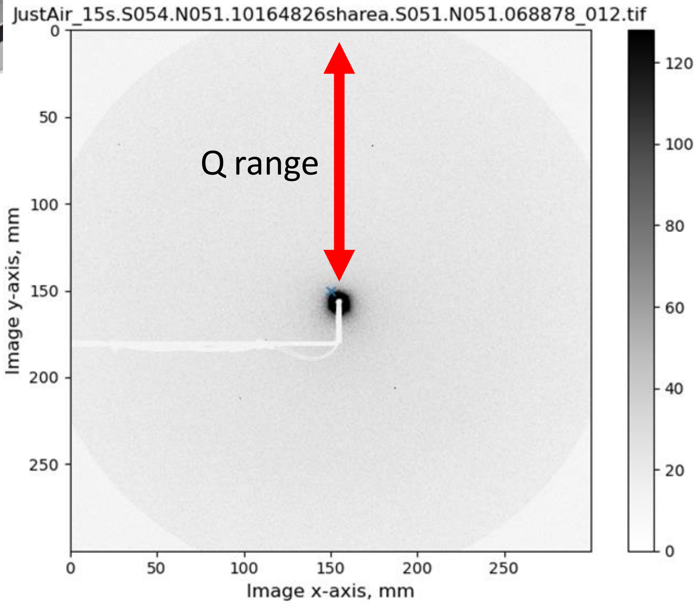
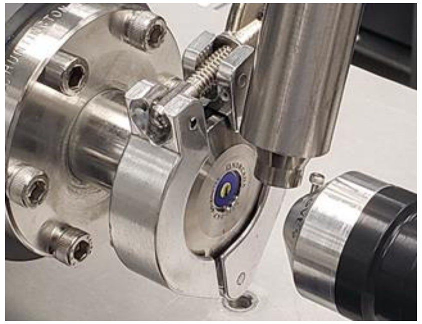
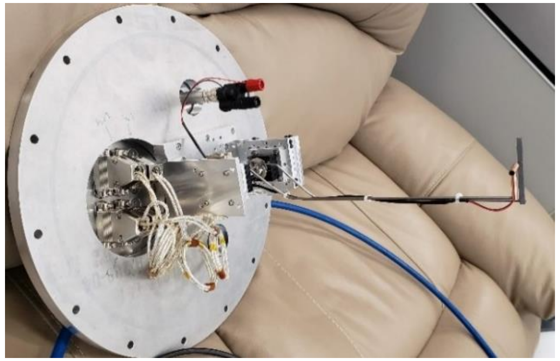


200 nm SiN window

3) Guard slit (0.80 mm)



Small air gap for sample (~35 mm)



Vacuum tube (≤ 10-2 Torr)
Beamstop with photodiode
-2.0 m
(4.0 mm)

13.5” Kapton window (125 μm)
~2100 pounds

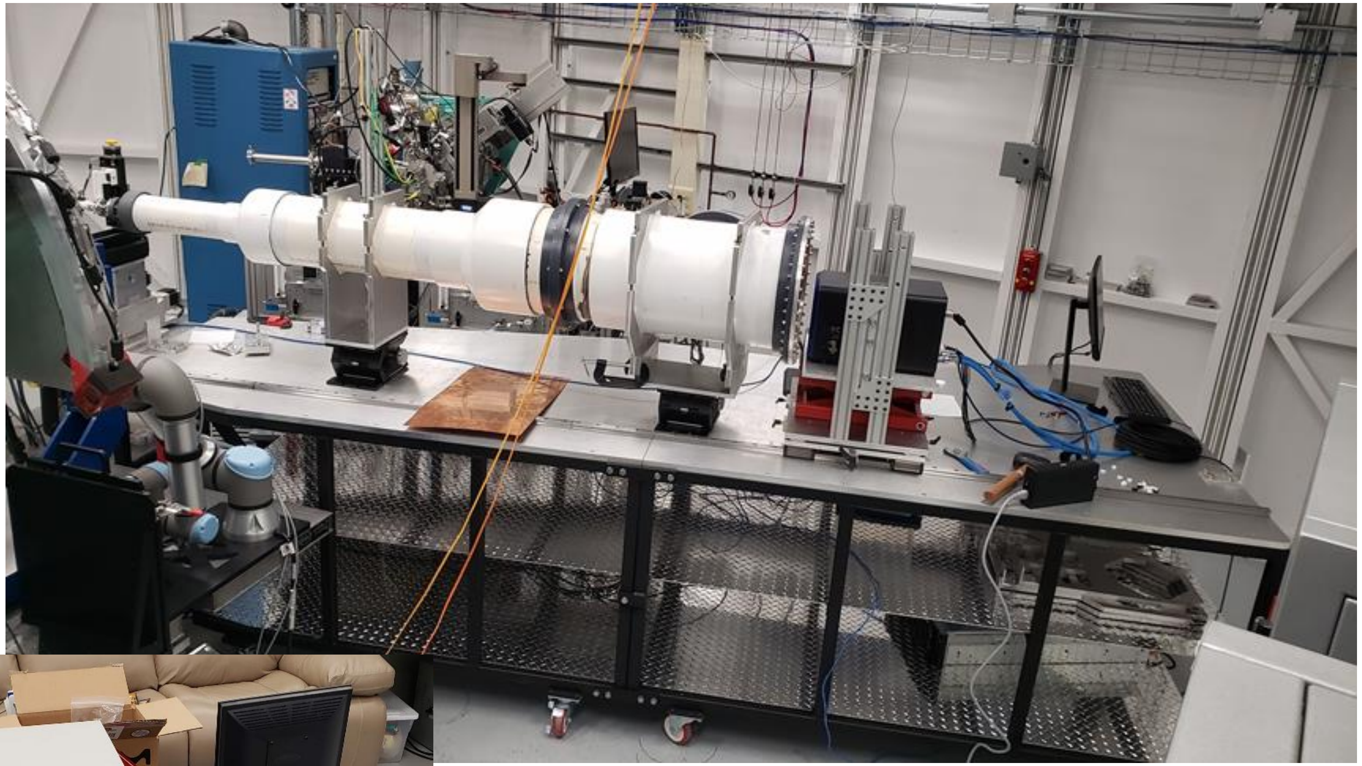
CLS SAXS instrument

New for next cycle (Jan-June 2025)

New table
- Easily go between SAXS and WAXS
- q_{min} from 0.005 Å^{-1} to 0.003 Å^{-1}

Optional Eiger 2 detector
- Strongly scattering samples
- Dynamic processes (sub-second)

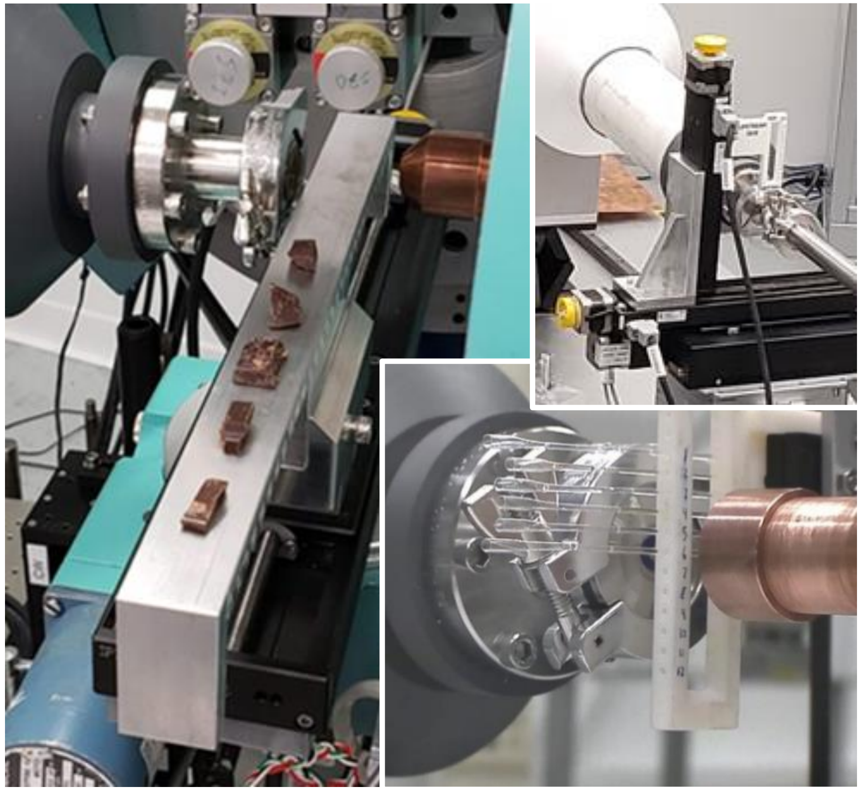
HPLC system
- Size exclusion chromatography-coupled SAXS



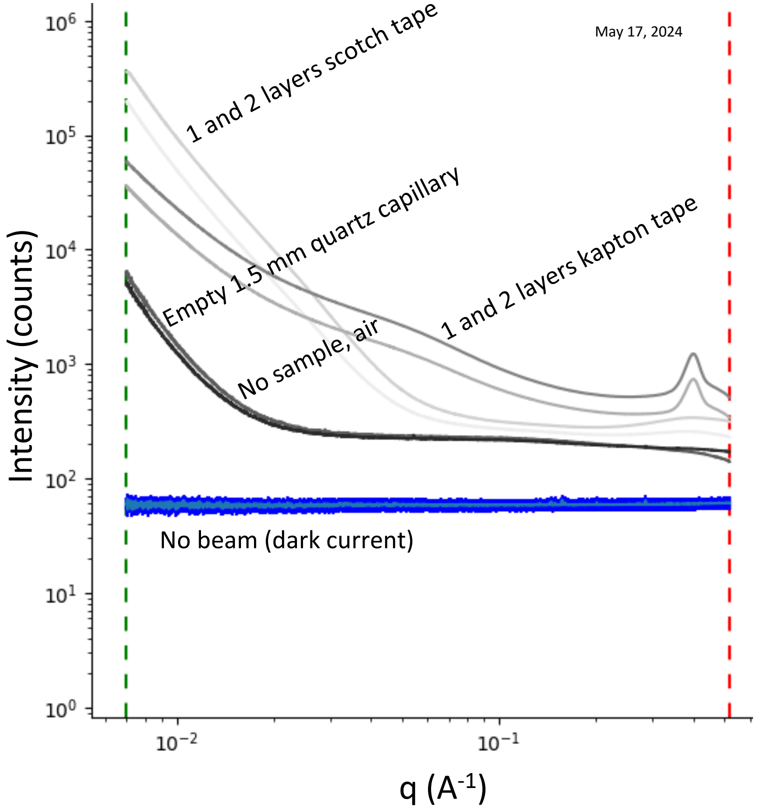
Sample preparation and mounting

Transmission

Freestanding, quartz capillaries, or on tape



One 30 s exposure for each, not background subtracted



Grazing incidence

Thin *flat* films on Si wafers, or glass slides
Min θ step = 0.01°

Calculate the transmission of your sample… 1/e is ideal
Bring blanks, Bring a range of concentrations
Bring a sample where you know what to expect
MUST buy your own capillaries

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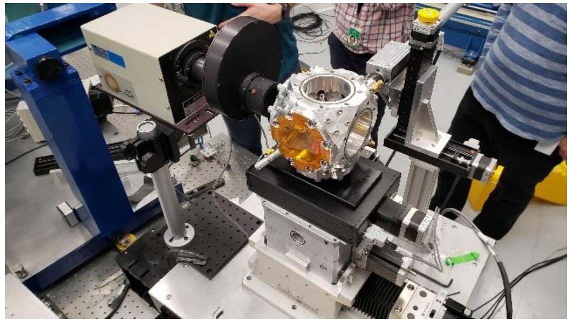
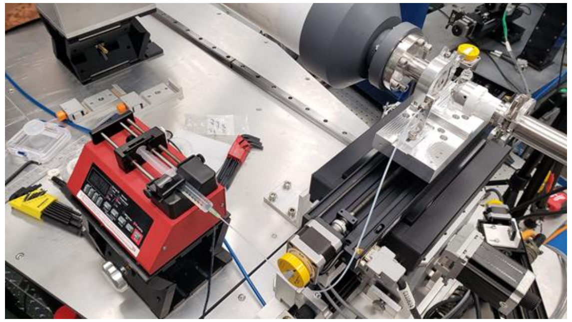
Sample preparation and mounting

We are flexible!

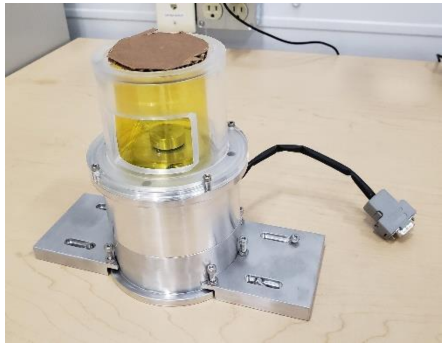
Operando solar cells
(sunlight, humidity, temperature)



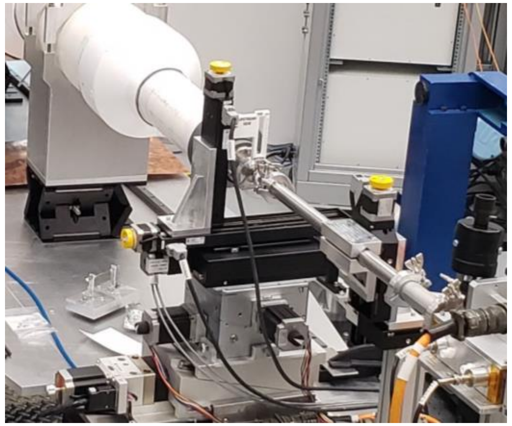
Liquid flow cell and syringe pump



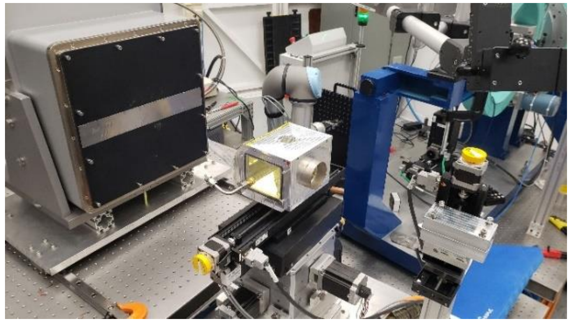
In-situ spin coater
(soon with heating)



Multi-sample transmission

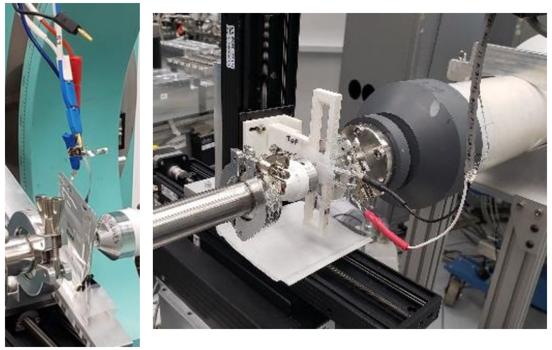


Helium box with Linkam heating and cooling
(-193 - +350°C)



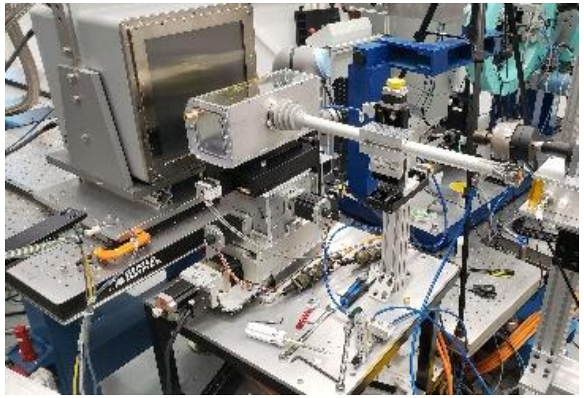
Oxford cryostream (-193 – +226°C)

Electric field/Electrochem SAXS

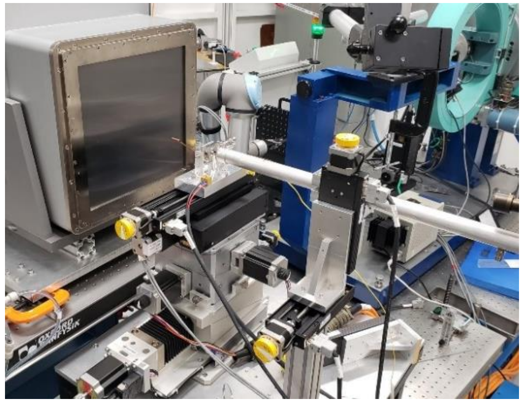


Automated multi-sample grazing

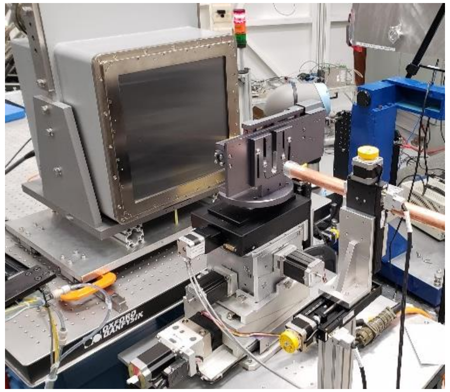
Automated multisample Helium box
for GIWAXS, also with mild heating



1000°C furnace with gas flow

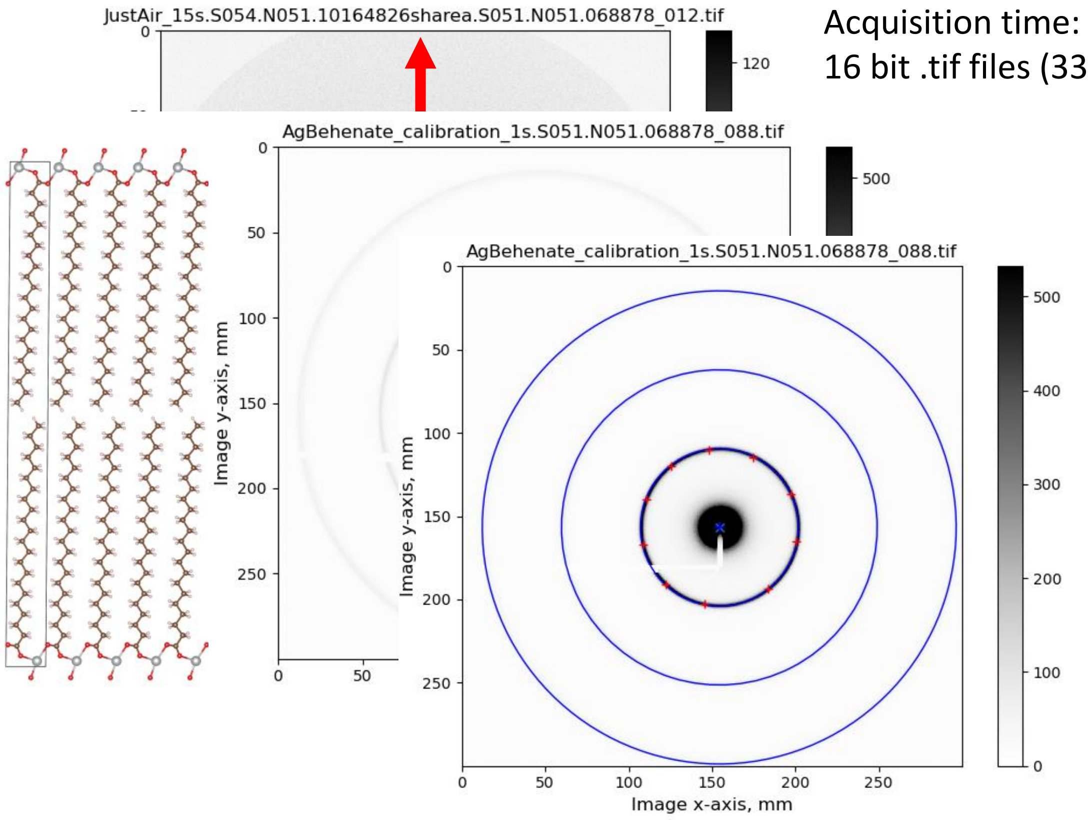


Stretching



Detector acquisition and calibration

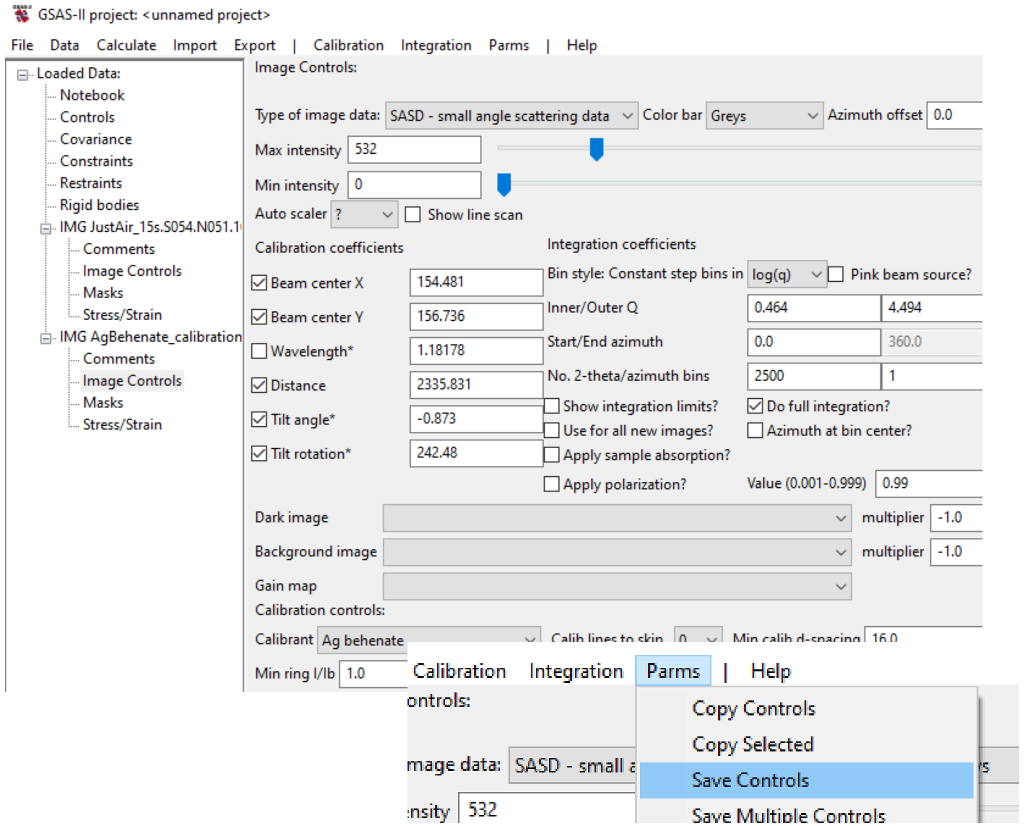
During your beamtime, note the wavelength, the approximate detector distance, and detector details



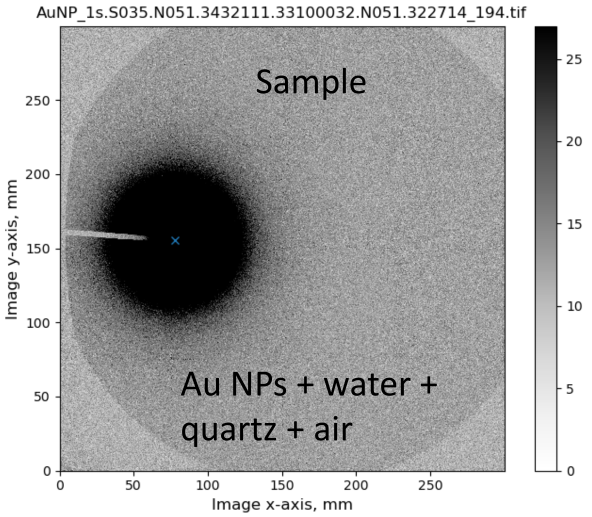
Acquisition time: 1 s - 90 s
16 bit .tif files (33 MB)

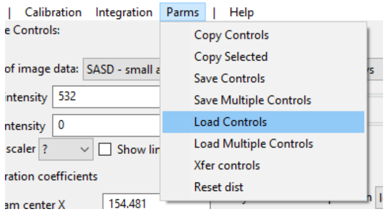
Ring	q (A-1)	d (nm)
1	0.1076	5.839
2	0.2152	2.920
3	0.3228	1.946
4	0.4304	1.460
5	0.5380	1.168

GSAS-II demo, "Calibration of an area detector"

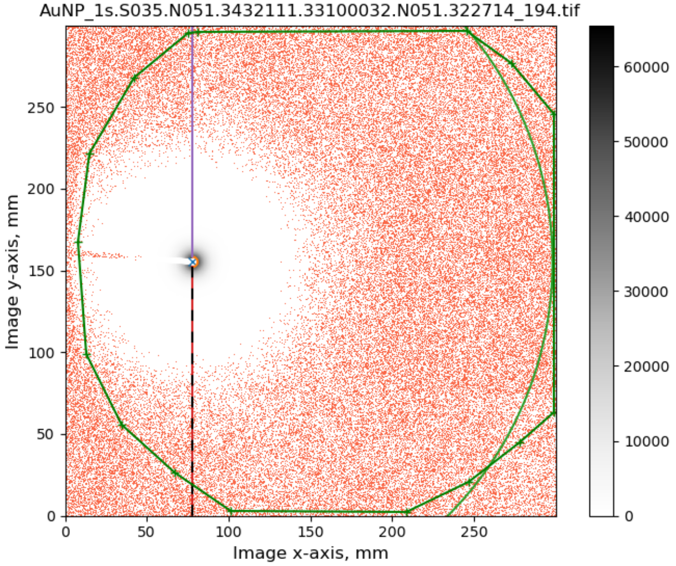


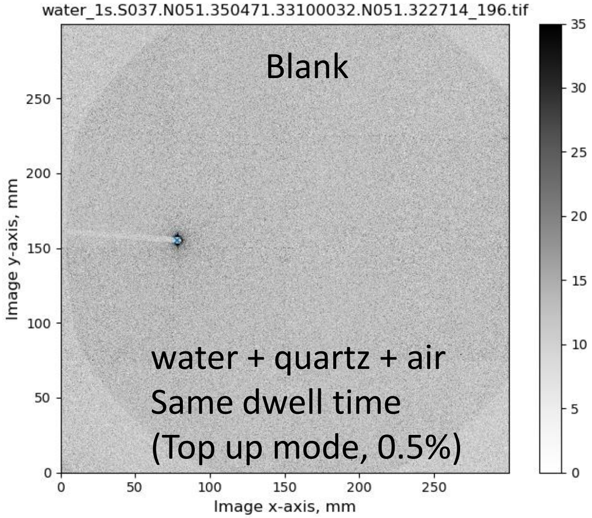
Data correction, and reduction to 1D plot

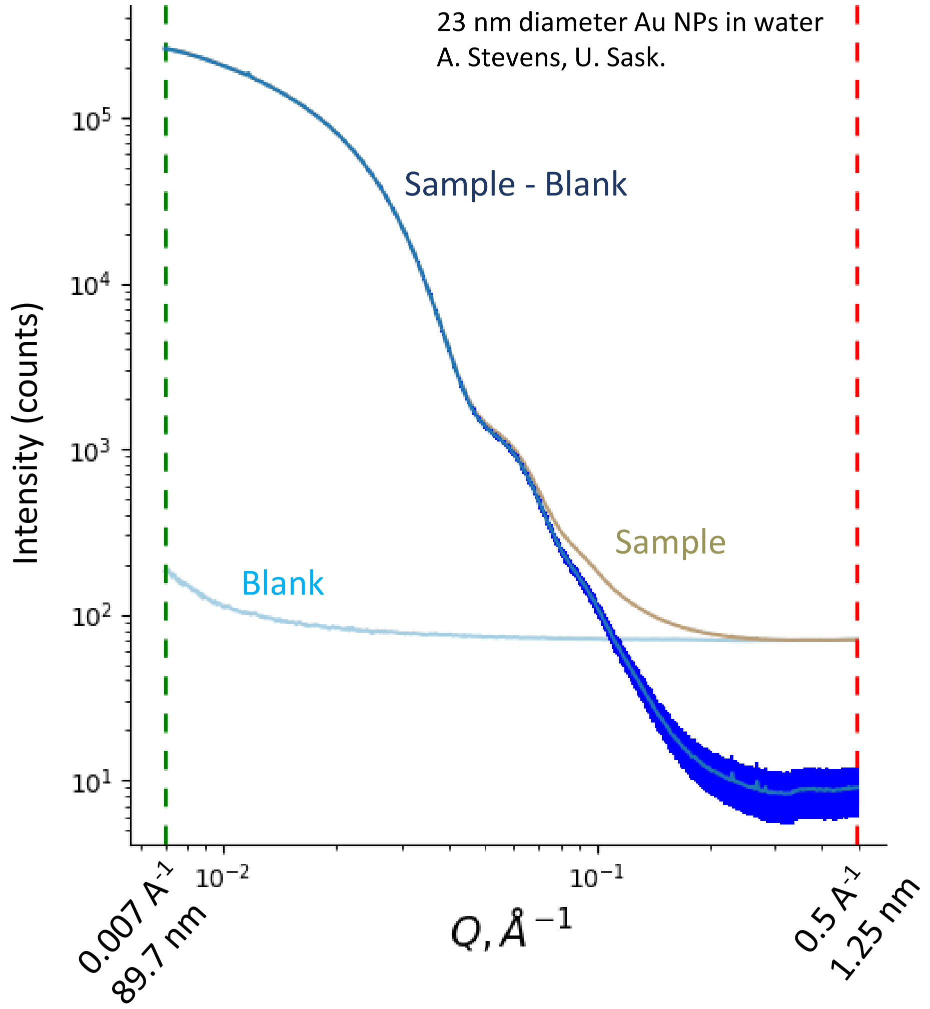




Subtract blank







Proper units of intensity are cm^{-1}
IF data are scaled to absolute intensity

5 tips for a great data collection experience

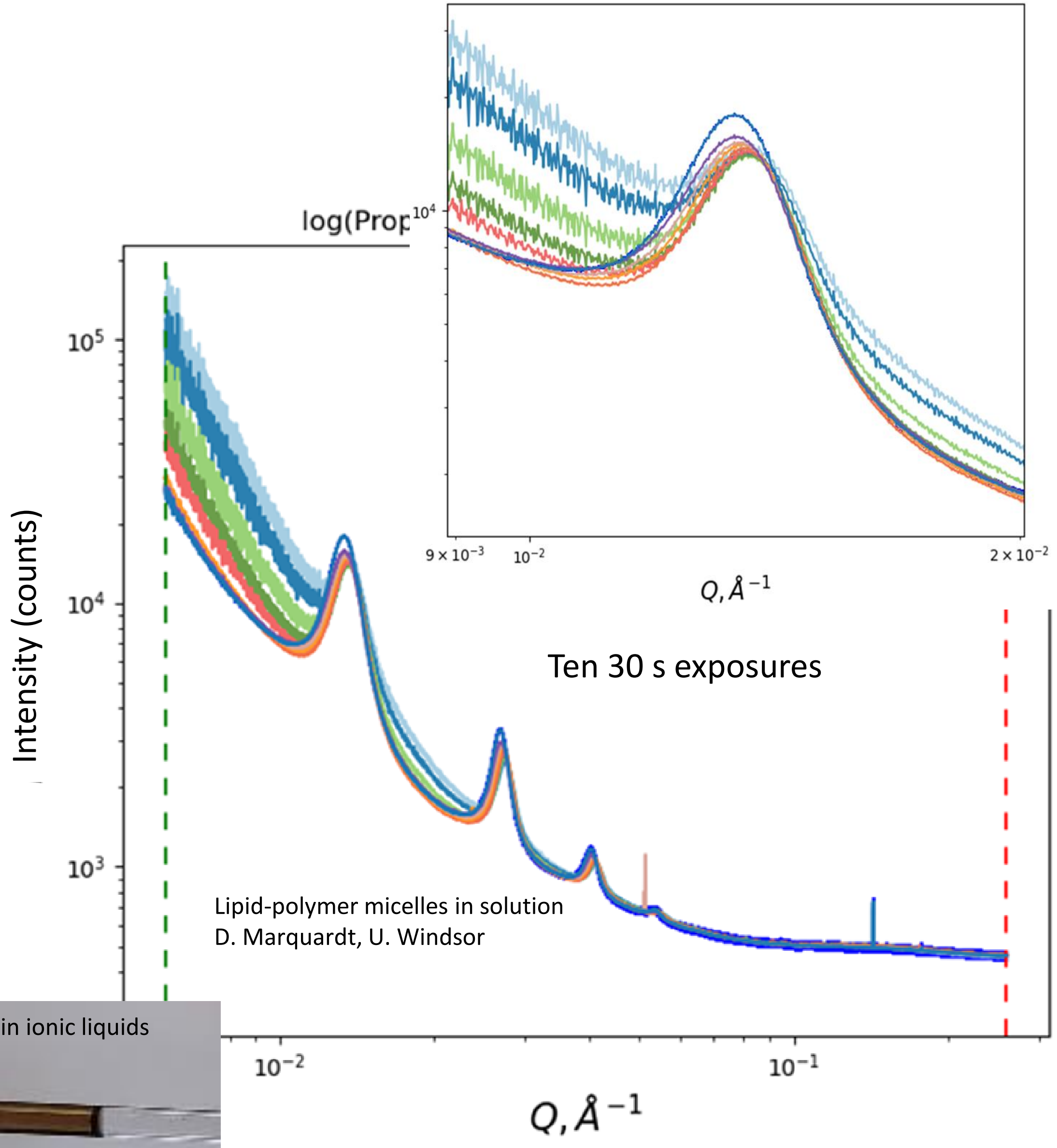
1) Know what q range you want, and include it in your proposal. We can tune the endstation to your problem.

2) Know what your data should look like. Find published examples, and bring a known good sample.

3) Get in contact ~2 weeks in advance.

4) Choose quality over quantity: Work up data as you go along, check for radiation damage at the start, many blank measurements.

5) It's your experiment.



Lipid-polymer micelles in solution
D. Marquardt, U. Windsor

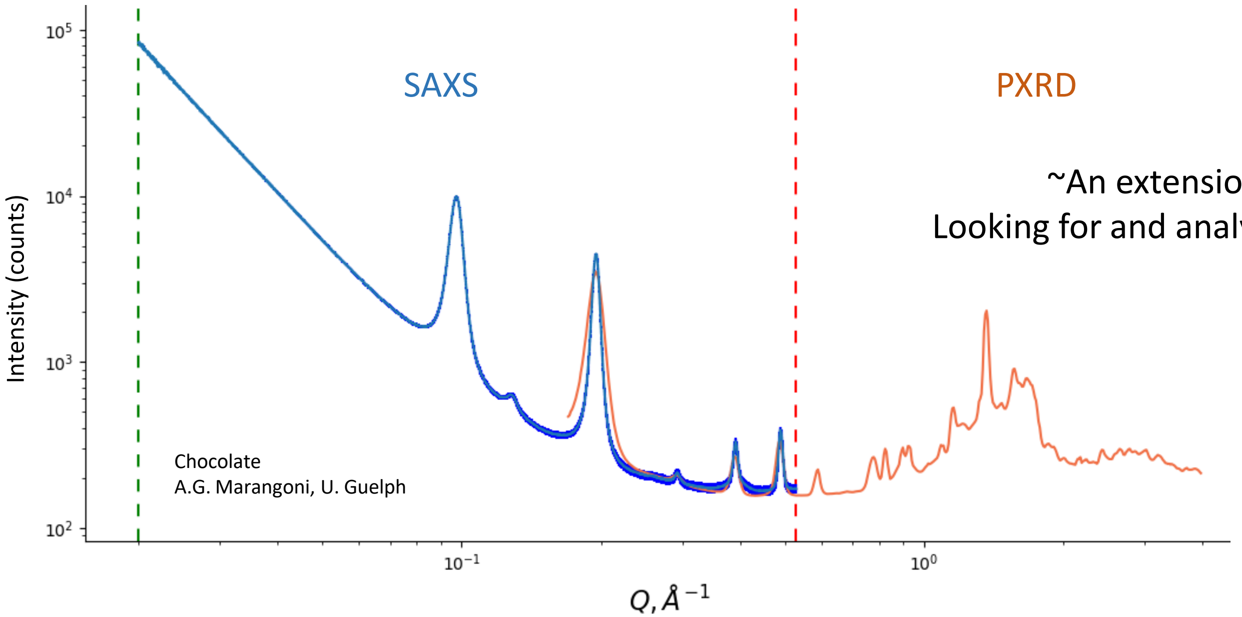

Pd nanoparticles in ionic liquids

Data analysis with examples

Origins of the SAXS signal

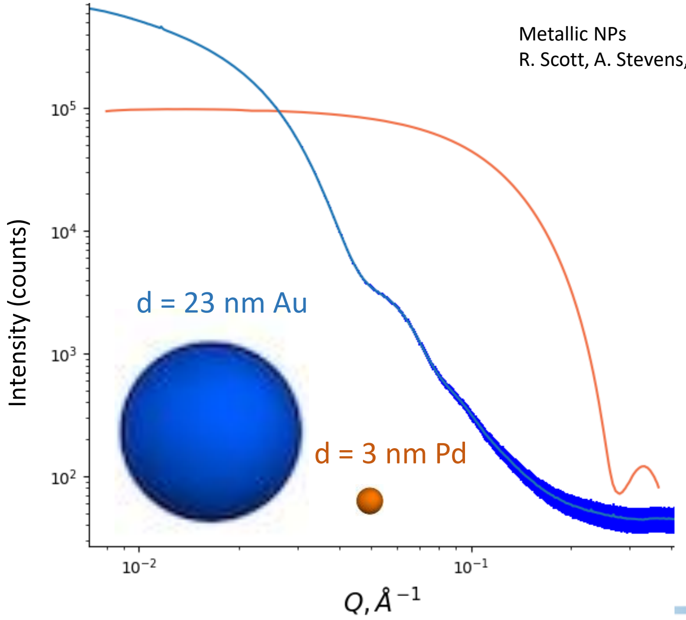
$$I(Q) = S(Q) \times P(Q)$$

S(Q) is the **structure** factor
- *Inter*-particle interferences
- High concentrations (>5% vol.)
- Ordering/packing of particles



~An extension of PXRD
Looking for and analyzing Bragg peaks

P(Q) is the **form** factor
- *Intra*-particle interferences
- Low concentration (dilute limit)
- Size/polydispersity/shape of particles



Analyzing the whole curve

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Phospholipid Self-Assembly in Cocoa Butter Provides a Crystallizing Surface for Seeding the Form V Polymorph in Chocolate

J.A. Stobbs, Alejandro G. Marangoni et al., *Cryst. Growth Des.* **24**, 7, 2685–2699 (2024)

Marangoni group
Dept. Food Science
University of Guelph

The nanostructure of chocolate is manipulated using time and energy-intensive "tempering"

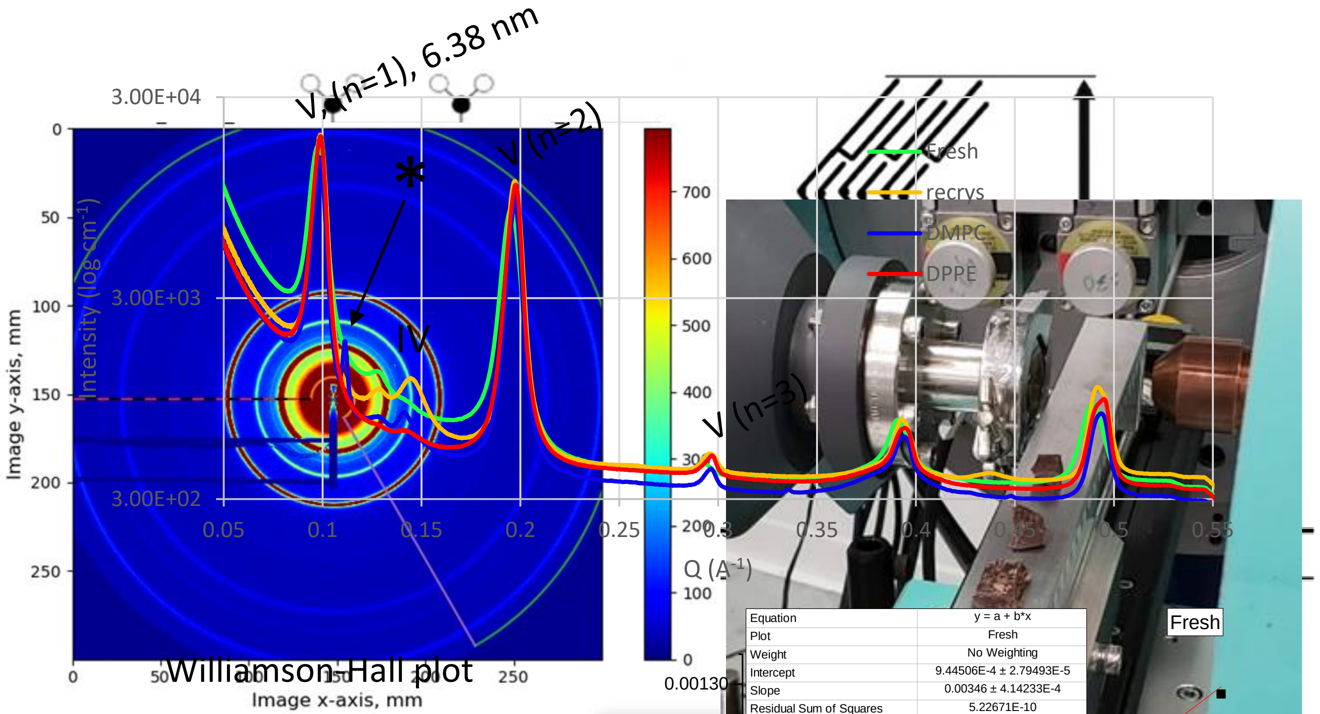
Guides crystallization of cocoa butter to "polymorph V"

Tempering of cocoa butter and chocolate using minor lipidic components

Jay Chen[1], Saeed M. Ghazani[1], Jarvis A. Stobbs[1,2] & Alejandro G. Marangoni[1]

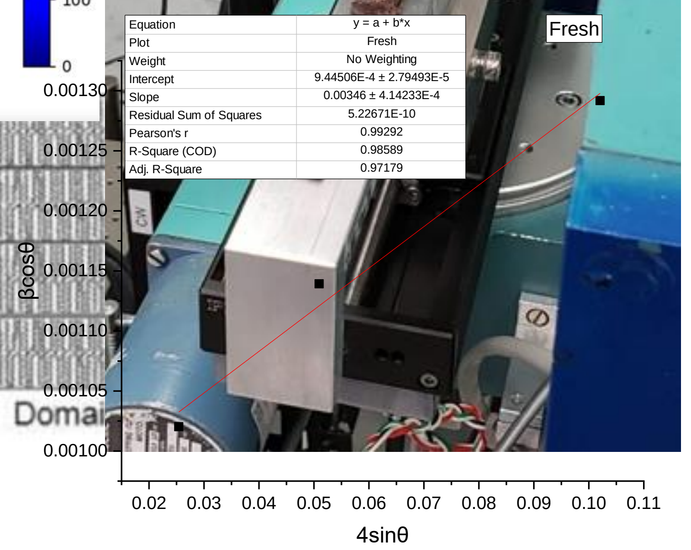
NATURE COMMUNICATIONS | (2021)12:5018

Why does it work?



Williamson-Hall plot

$$B\times\cos(\theta)=\underbrace{\frac{K\times\lambda}{Size}}_{\text{y-intercept}}+\underbrace{4\times Strain\times\sin(\theta)}_{\text{slope}}$$



crystallite size and strain from peak width (*B*) analysis

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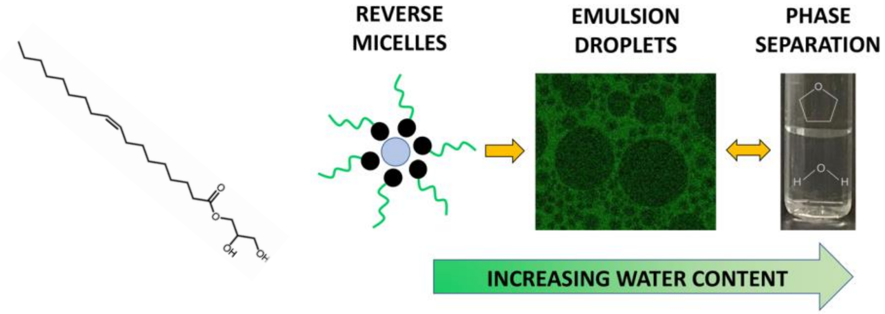
Environmental science: Waste water treatment

Pensini group
School of Engineering
University of Guelph

Scientific Reports **12**, 15832 (2022)
J. Molecular Liquids **367**, 120551 (2022)
Physics of Fluids **34**, 097119 (2022)

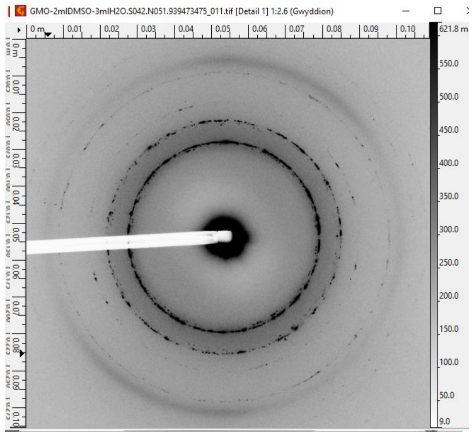
Separating solvents (THF, DMSO, DMF and acetonitrile) and metals from waste water using amphiphiles, emulsifiers and surfactants



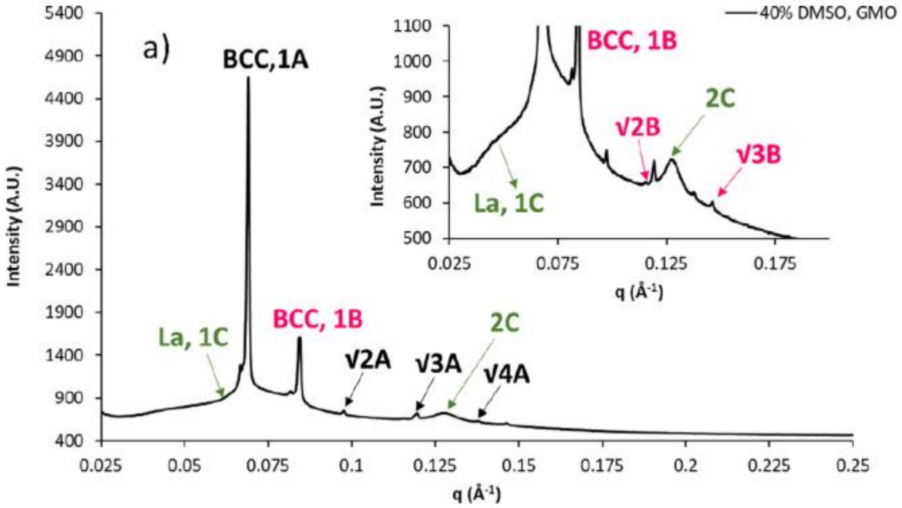
Mechanism of solvent separation?

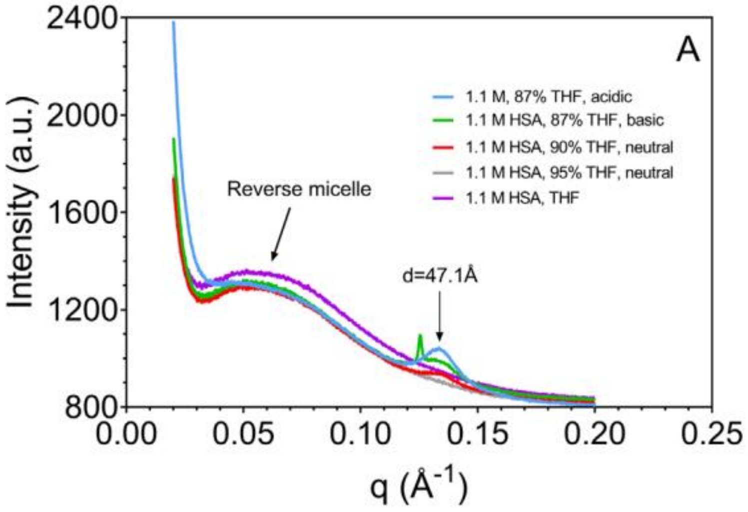
	1.1 M HSA, 87% THF, acidic pH	ξ (Å)	d (Å)	d/x
a_2	1.158	19.5	86.0	4.4
c_1	−98.92			
c_2	18 245			

Powder diffraction ... ??

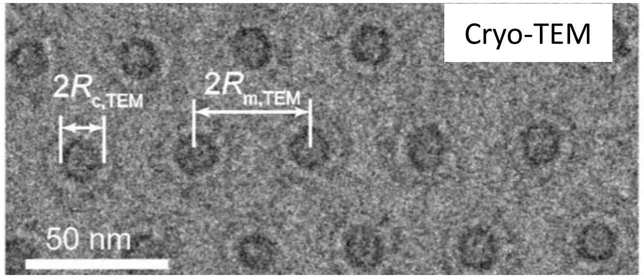


Symmetry	Peak position ratio
BCC (Im3m)	√2, √3, √4 …





Reverse micelle lattice



L. Chen et al., *PNAS* **115**, 7218-7223 (2018)

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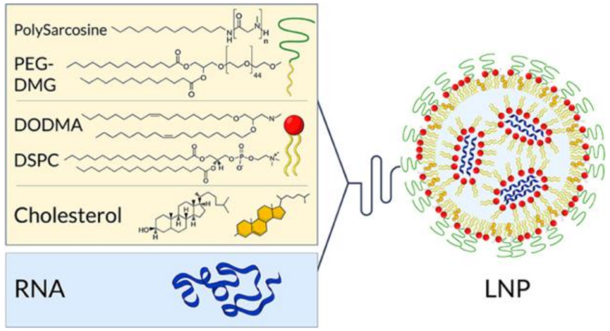
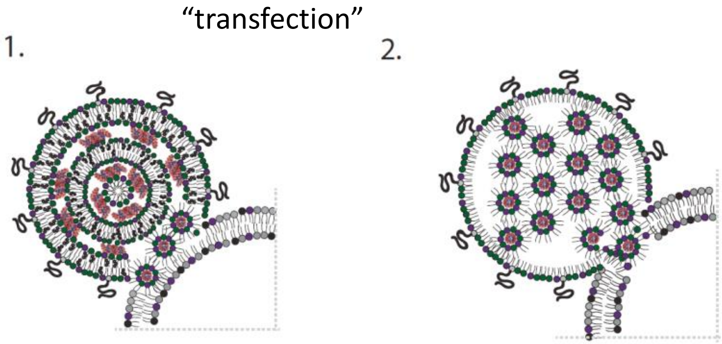
Pharmacy: Liposomes for RNA delivery

Pure mRNA degraded by enzymes in tissues
Hide it inside a liposome NP to get it into cells



ACS Appl. Nano Mater. 2020, 3, 11, 10634–10645

Pfizer, Moderna COVID-19 mRNA vaccines

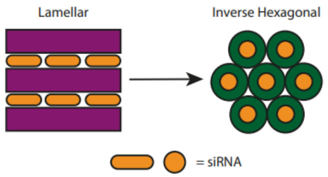


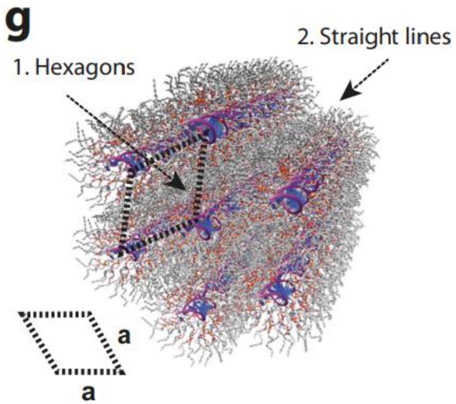
In situ L_α to H_{II} transition

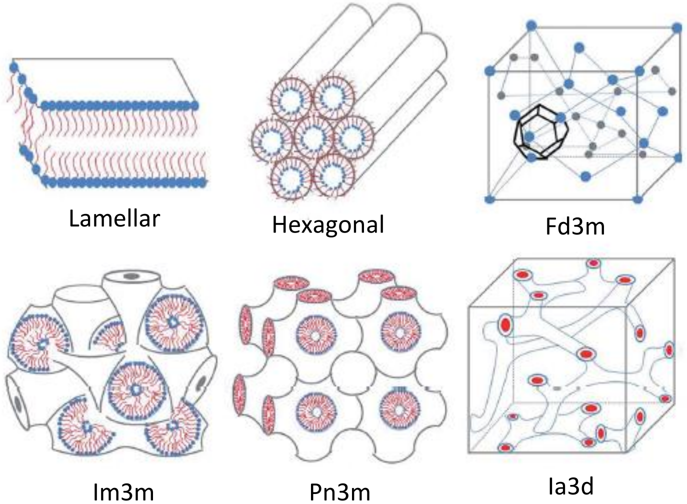
Pre-programmed H_{II} remains intact

Nature Comm. 15, 1303 (2024)

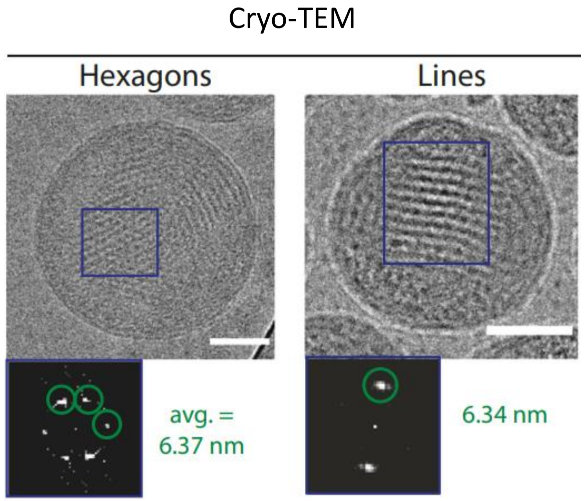
Lipid phase affects transfection efficiency



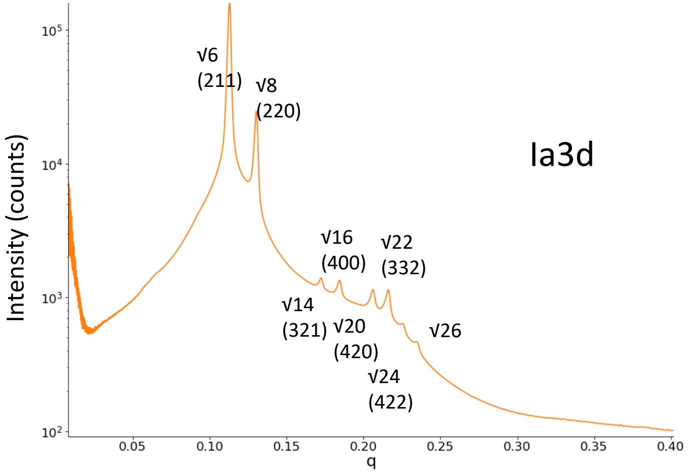




Y. Huang, S. Gui, RSC Adv., 2018, 8, 6978–6987



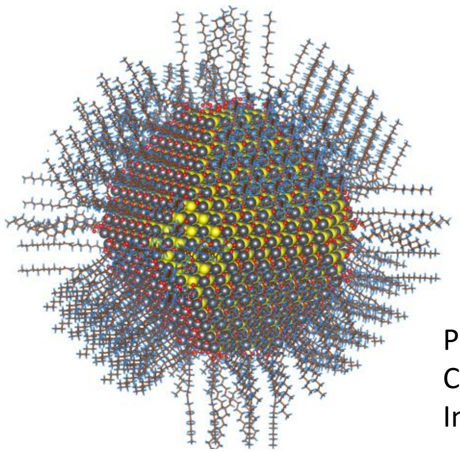
Nature Comm. 15, 1303 (2024)



Types of LLC phase	SAXS
Lamellar phase	1 : 2 : 3 : 4
Im3m	$\sqrt{2} : \sqrt{4} : \sqrt{6} : \sqrt{8}$
Pn3m	$\sqrt{2} : \sqrt{3} : \sqrt{4} : \sqrt{6} : \sqrt{8} : \sqrt{9}$
Ia3d	$\sqrt{6} : \sqrt{8} : \sqrt{14} : \sqrt{16} : \sqrt{20} : \sqrt{22}$
Fd3m	$\sqrt{3} : \sqrt{8} : \sqrt{11} : \sqrt{16} : \sqrt{19}$
Reverse hexagonal	$1 : \sqrt{3} : 2 : \sqrt{7}$

Energy materials: Quantum dot solar cells

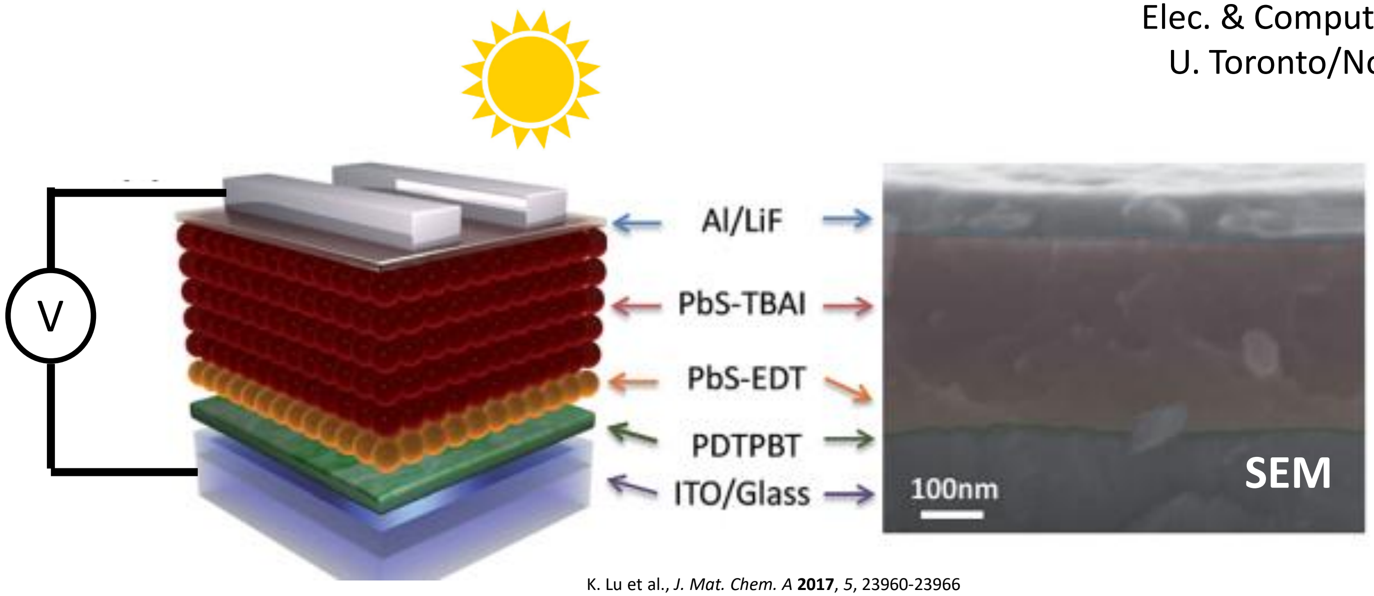
Sargent group
Elec. & Computer Engineering
U. Toronto/Northwestern U.



PbS
$CsPb(Br/I)_3$
InAs

https://www.olcf.ornl.gov/2015/05/05/demystifying-quantum-dot-conundrums/

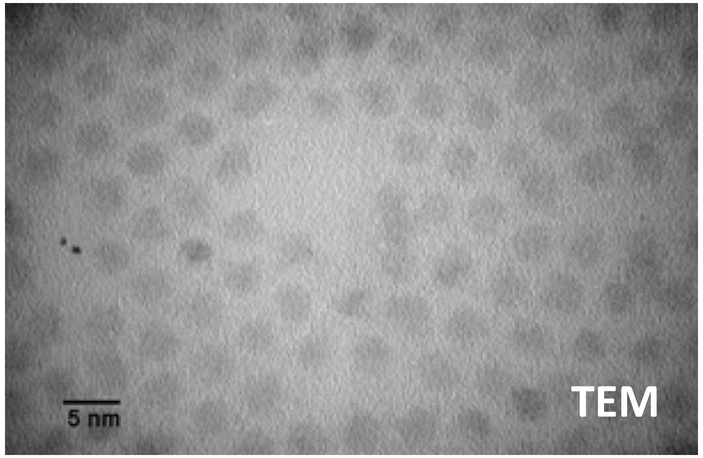
Efficient exciton generation
Size-based, tune-able band gap



K. Lu et al., *J. Mat. Chem. A* **2017**, *5*, 23960-23966

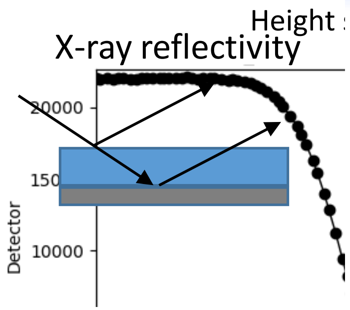
QD films deposited layer-by-layer using spin coating
Top electrodes deposited using thermal evaporation

18.1% record efficiency in 2022
Performance improved by tuning:
- Particle size distribution or polydispersity
- How particles pack and the interparticle distance



M. Yuan et al., Adv. Mater. 2014, 26, 3513–3519

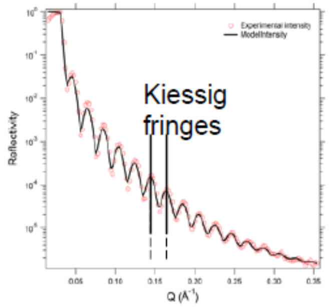
Control the size and packing by:
- Ligand exchanges cycles, centrifuge to remove largest QDs
- Rinsing steps during buildup
- Spin speeds, solvents, additives

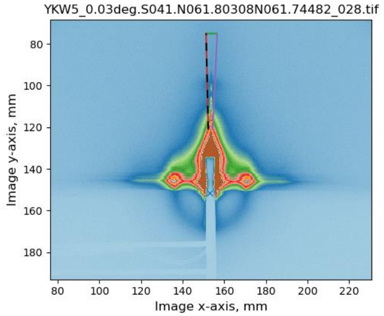


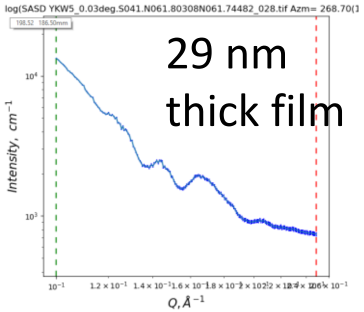
Simple estimation of film thickness

Fringes with uniform spacing

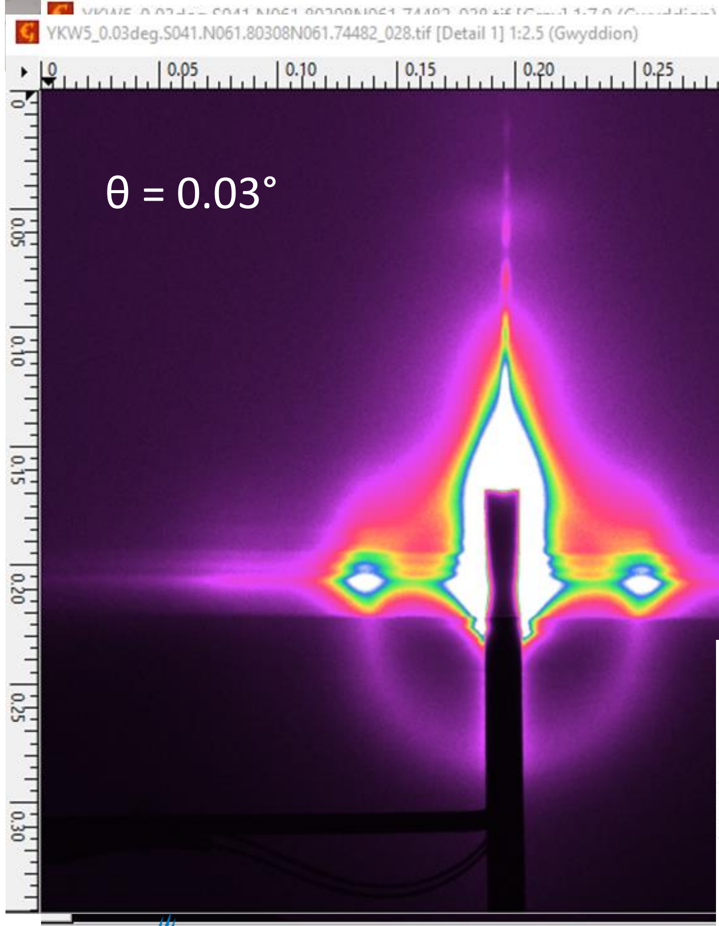
Thickness of the layer : $t = \dfrac{2\pi}{\Delta q_z}$

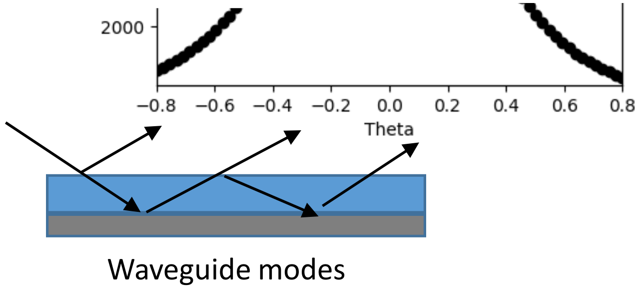






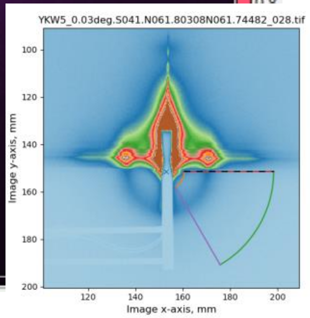
- Reflectivity gives film thickness

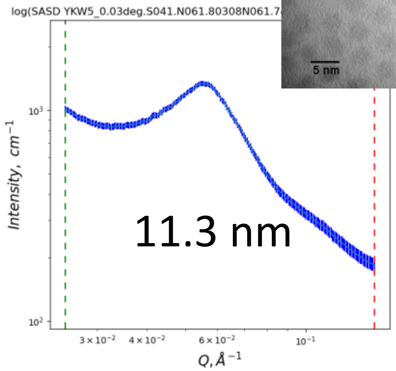


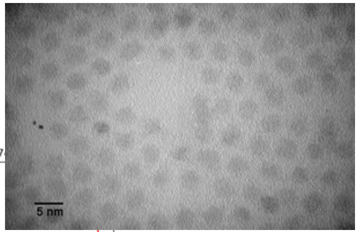


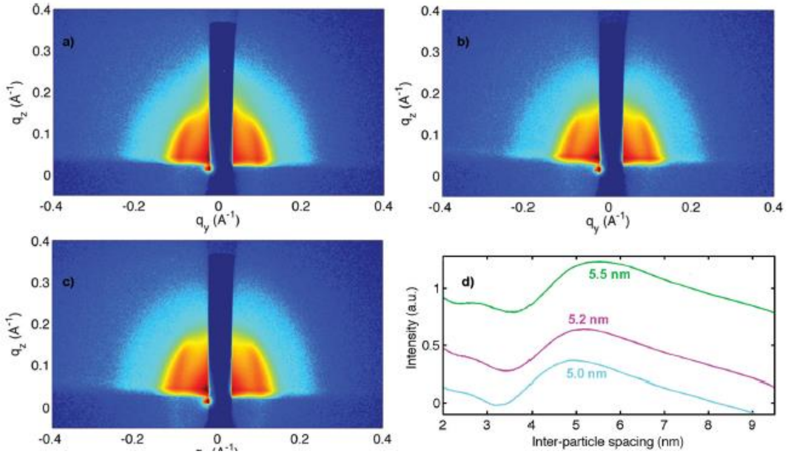
- Position gives interparticle distance
- Peak width gives size distribution
- Average over relatively huge area

- Indicates very flat, high quality film
- Adjusting grazing angle can give depth information









M. Yuan et al., Adv. Mater. 2014, 26, 3513–3519

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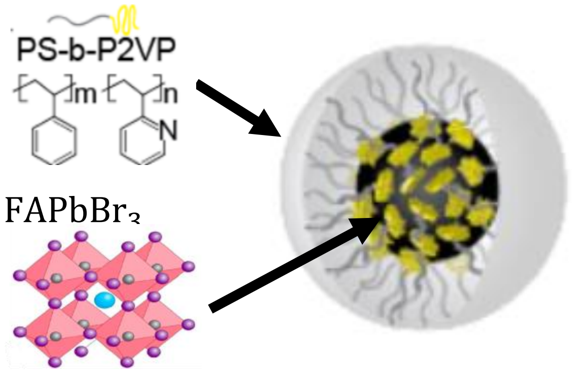
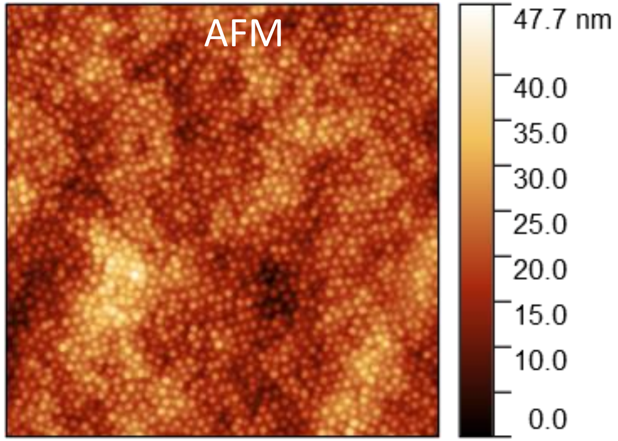
GISAXS - Lab X-ray source vs. Synchrotron

Turak group
Concordia, Physics

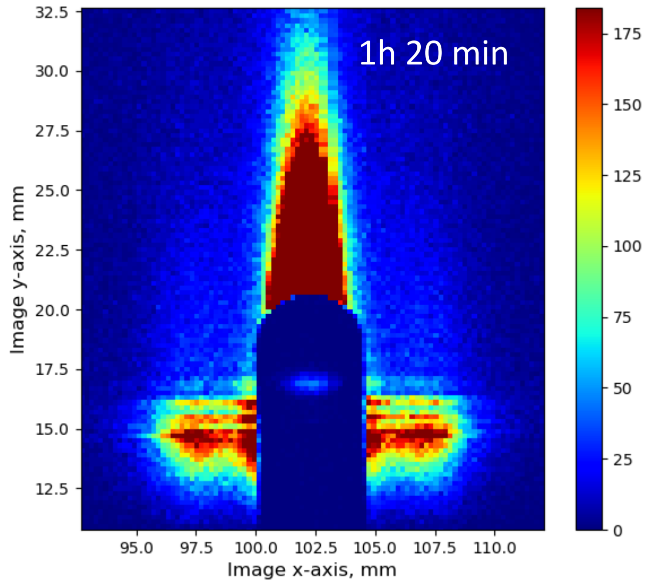
TURAK FUNCTIONAL NANOMATERIALS RESEARCH GROUP
ayse.turak@concordia.ca



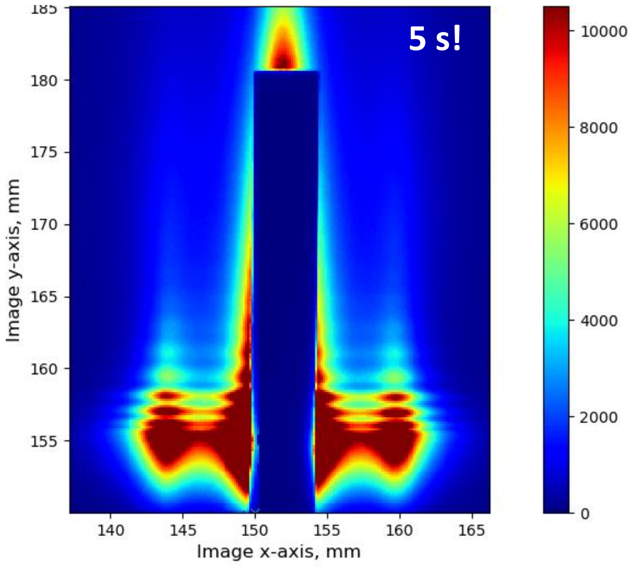


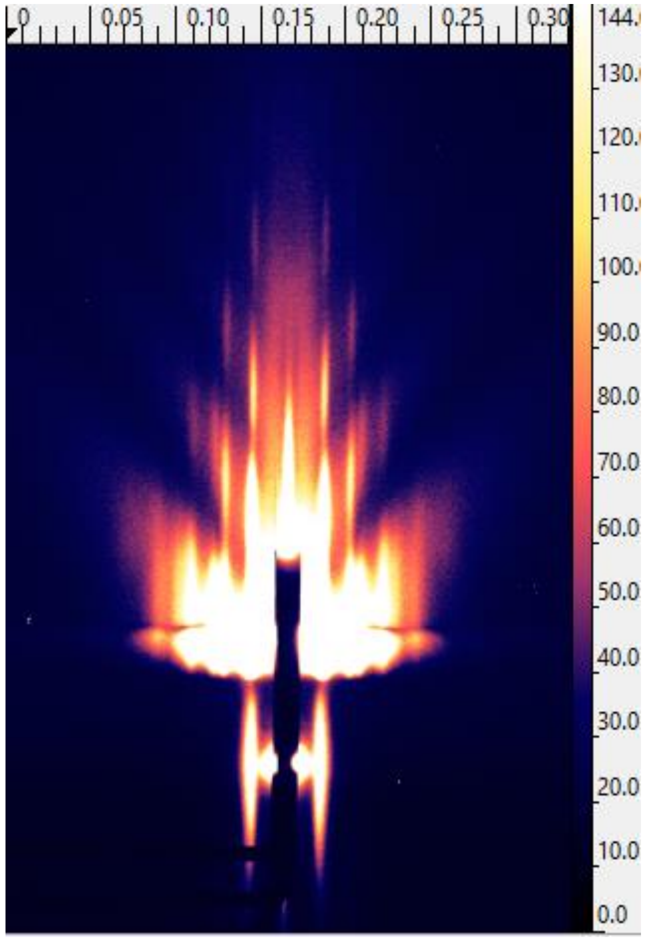
- Horizontal stripes indicate stacked layers of NPs, layer spacing.
- Vertical stripes indicate lateral ordering of the NPs, lateral distance.
- Peak width indicates size distribution and disorder in the system.

Lab (McGill, Anton Parr)



Synchrotron (WLE)





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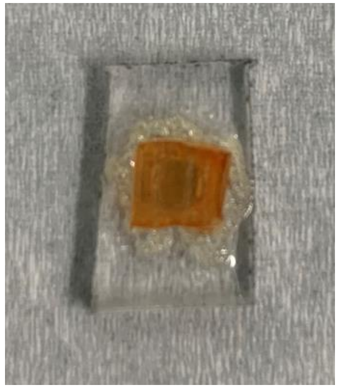
$I(Q) = S(Q) \times P(Q)$

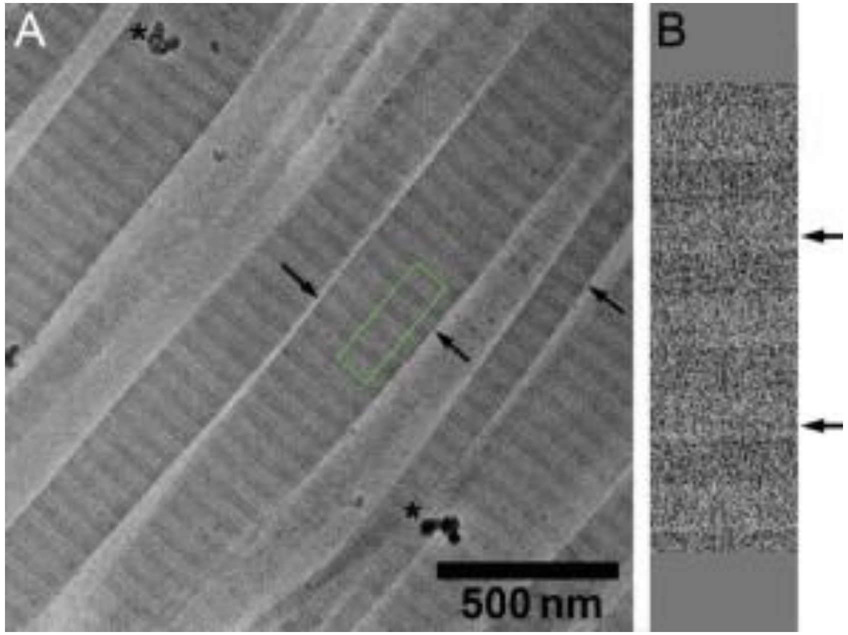
S(Q) is the **structure** factor
- *Inter*-particle interferences
- High concentrations (>5% vol.)
- Ordering/packing of particles

~An extension of PXRD
Looking for and analyzing Bragg peaks

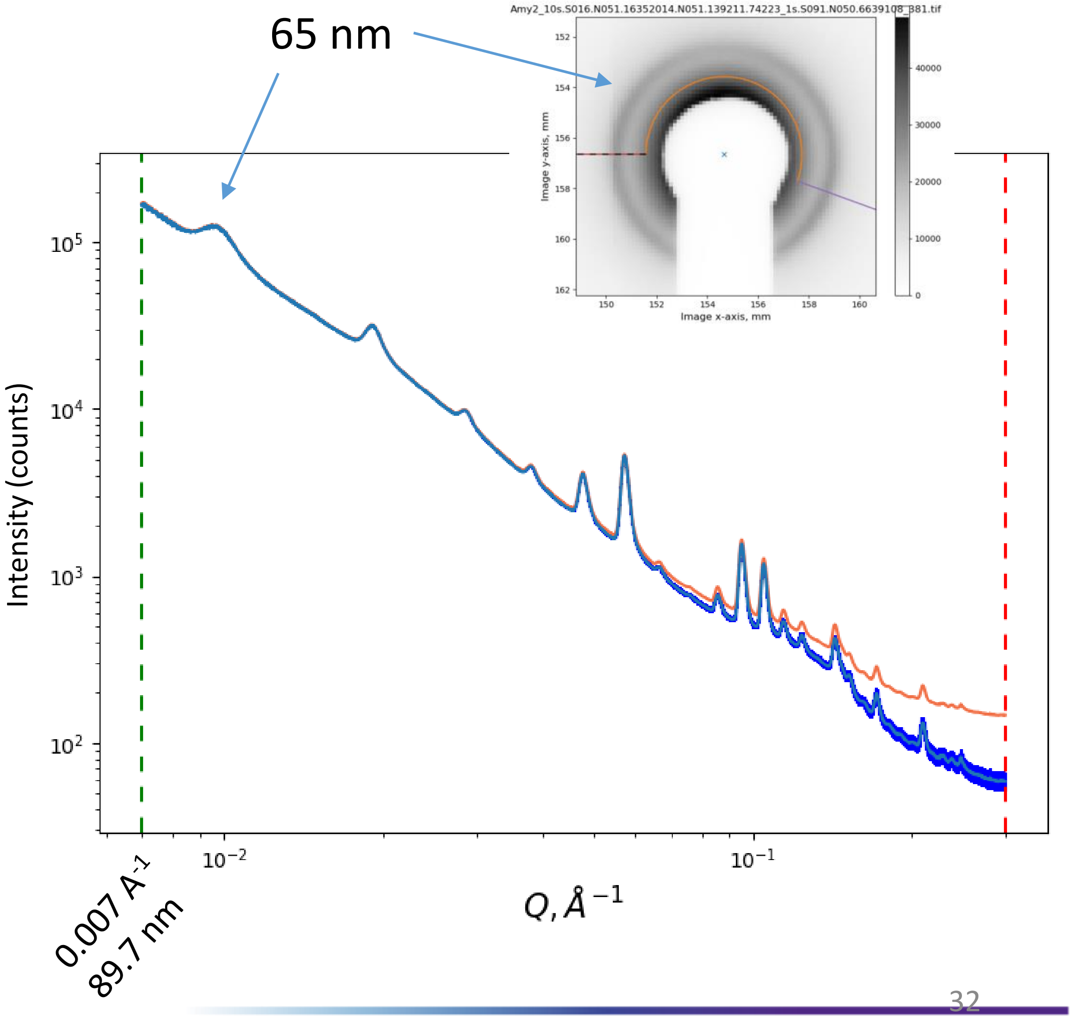


Sea cucumber dermis
M. Harrington, McGill

Type 1 collagen fibril gaps



https://doi.org/10.1016/B978-0-12-416617-2.00009-6



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Origins of the SAXS signal

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P(Q) is the **form** factor
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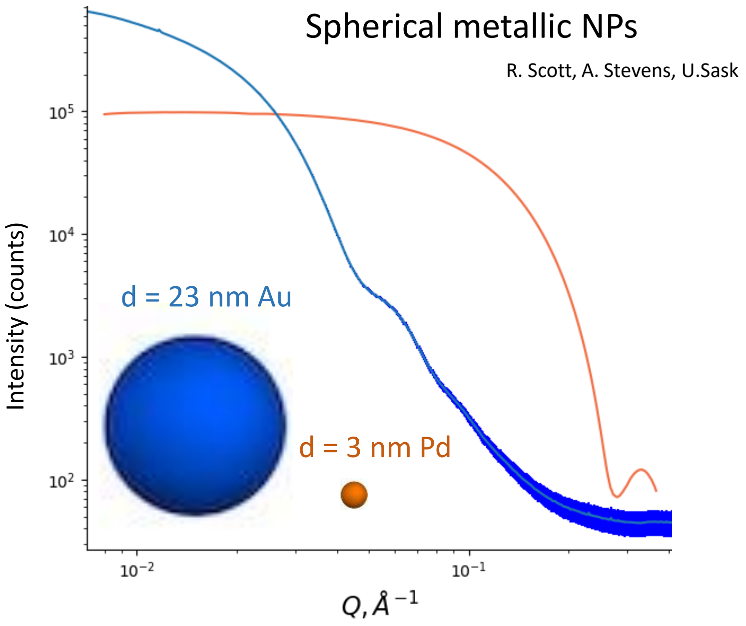
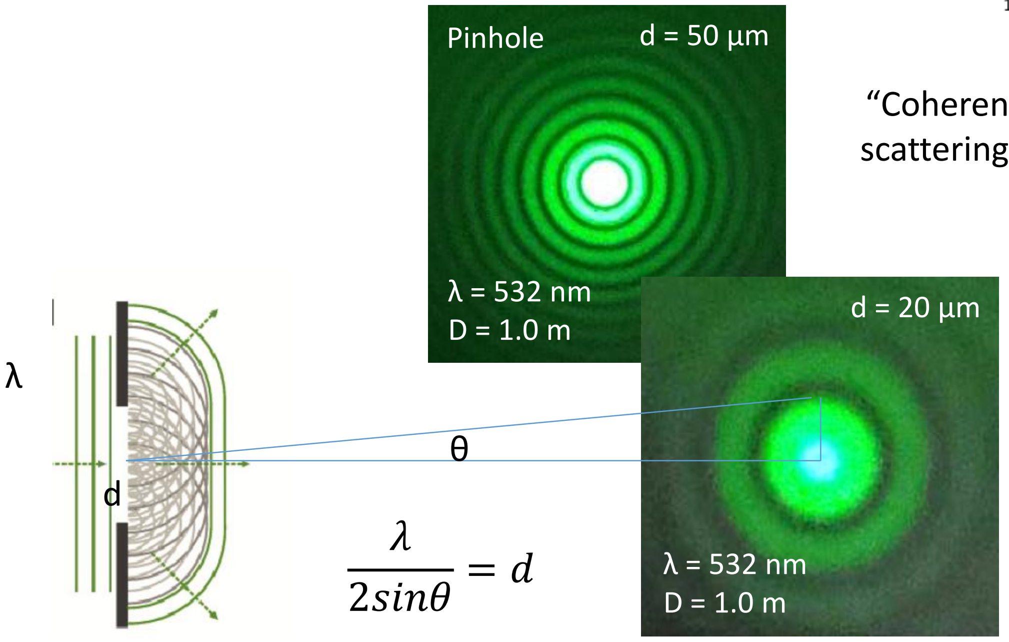
- Size, shape, polydispersity of particles



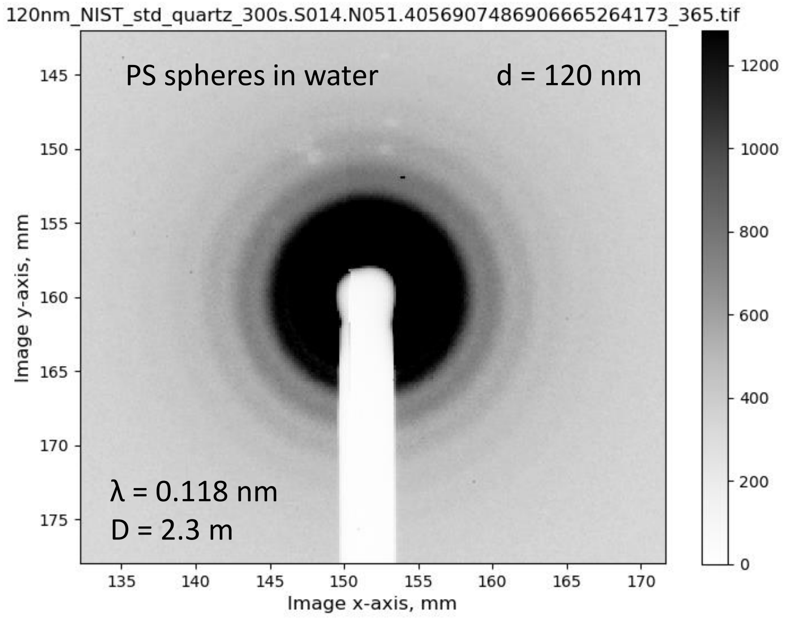
Sample prep is critical !!!

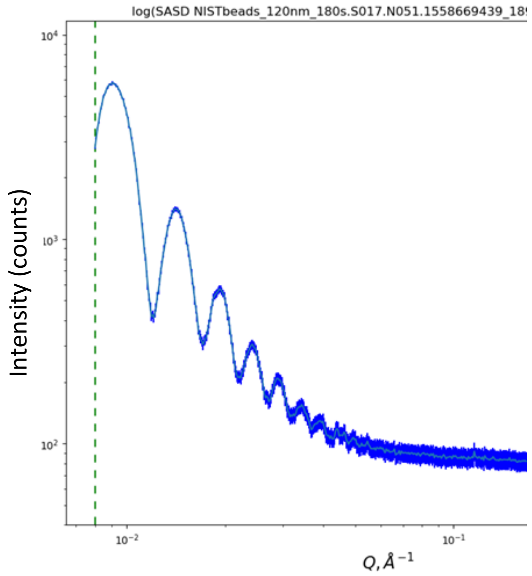
1. Dilute (~1 – 10 mg/mL) no interparticle interactions
2. Pure
3. Monodisperse



"Coherent scattering"

$$\frac{\lambda}{2sin\theta} = d$$





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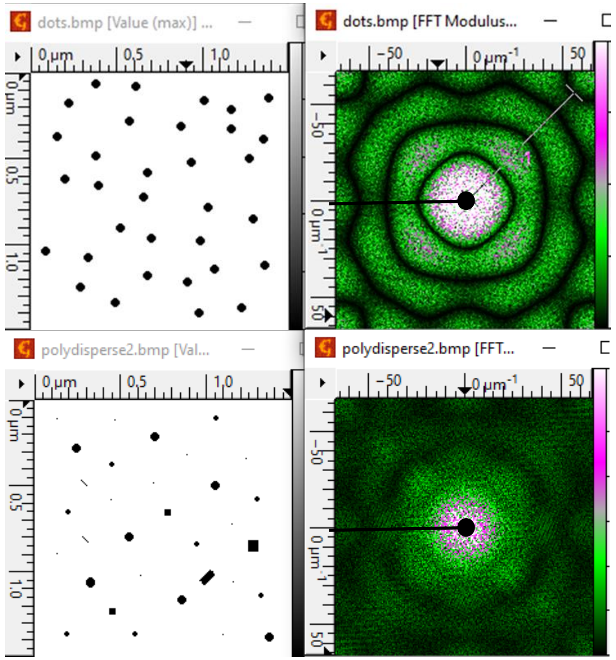
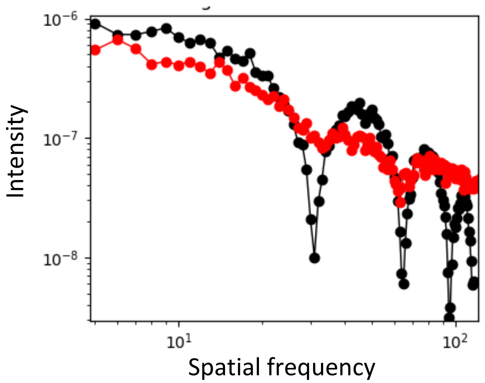
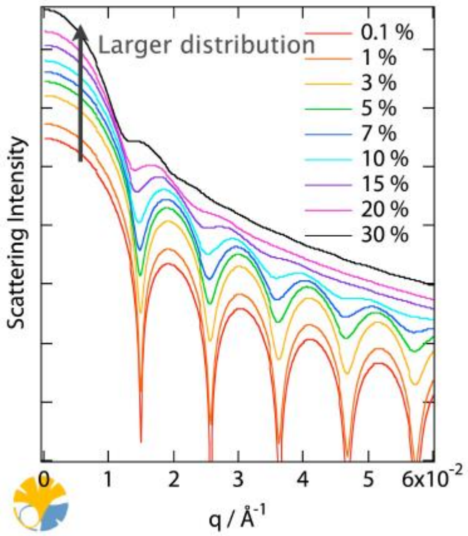
But there are limits!

Solutions must be monodisperse, and pure

Simulated sample → FFT → Simulated SAXS detector image



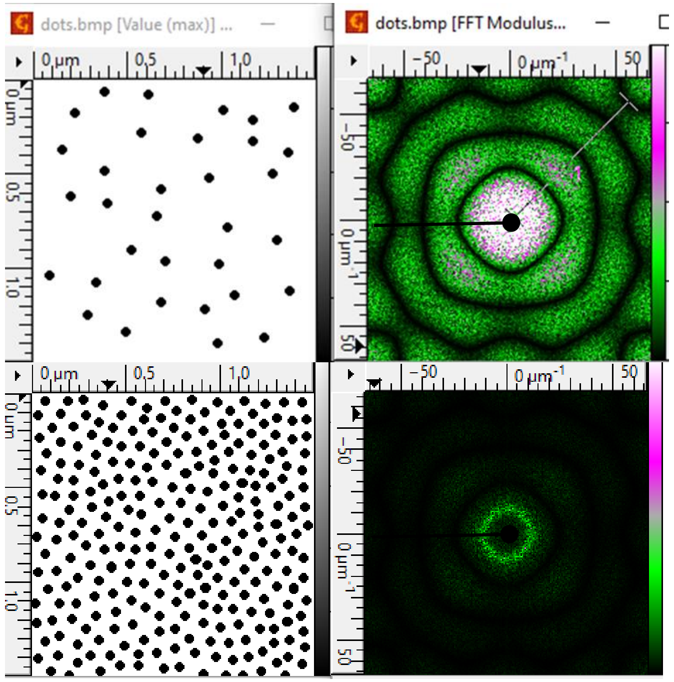


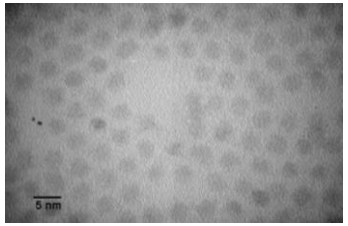


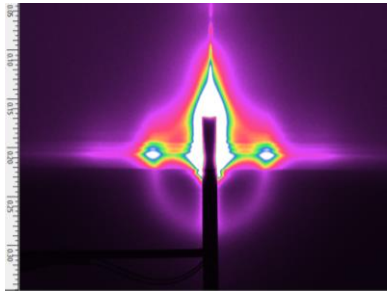
Form factor oscillations smear out when size polydispersity is ≥15%

Solutions must be dilute: No interparticle effects

~1 – 10 mg/mL… Bring many concentrations







M. Yuan et al., Adv. Mater. 2014, 26, 3513–3519

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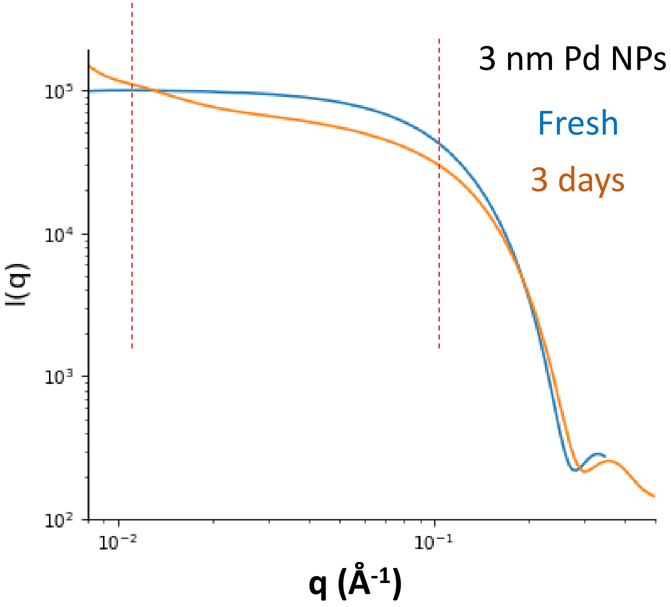
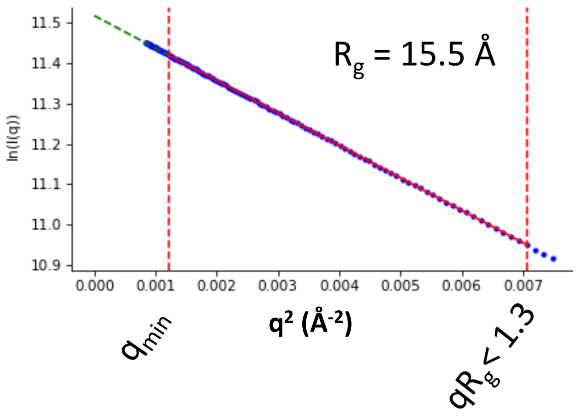
Form factor data analysis: Catalytic nanoparticles

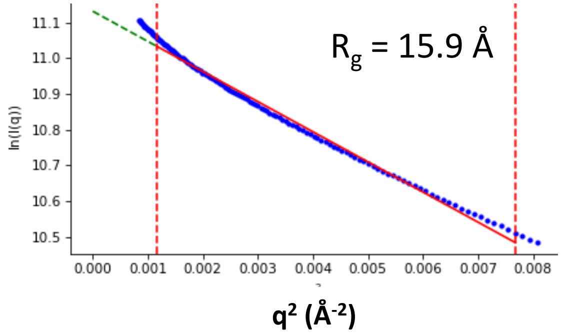
Guinier's approximation: $$I(q) \approx I(0)e^{-q^2 R_g^2/3}$$

R_g: Radius of gyration
I(0): Intensity at q=0



Guinier plot

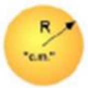




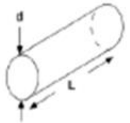
R_g is the square root of the average squared distance of each scatterer from the particle center

RMS distance of the objects parts from its center of mass

➢ Particle radius



Sphere $R_g^2 = \frac{3}{5}R^2$ Thin rod $R_g{}^2 = \frac{L^2}{12}$ Thin disc $R_g{}^2 = \frac{R^2}{2}$ Cylinder $R_g{}^2 = \frac{d^2}{8} + \frac{L^2}{12}$



q range required

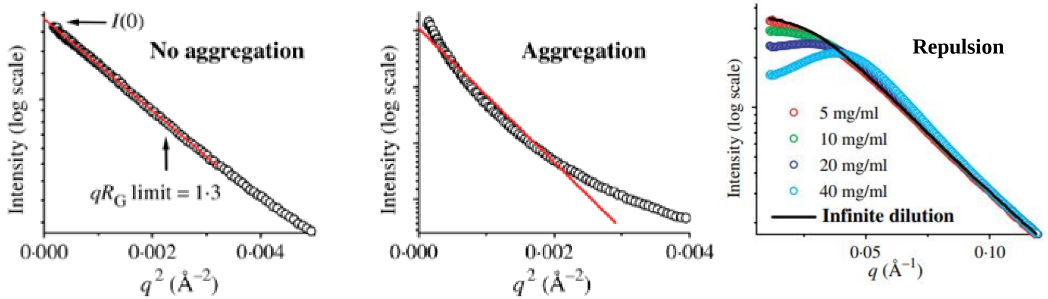
Globular, disc: $q_{min}{}^*R_g$ <1.0 to ~1.3
Linear: $q_{min}{}^*R_g$ <0.65 to ~1.0

If q_{min}= 0.005 A^{-1}

$R_g$max = 20 nm
$R_g$max = 13 nm

https://bioxtas-raw.readthedocs.io/en/latest/saxs/saxs_guinier.html
D. Putnam et al. Quart. Rev. Biophys. 40, 191-285 (2007)

➢ Sample quality: non-linearity can indicate aggregation, repulsion, radiation damage



D. Putnam et al. Quart. Rev. Biophys. 40, 191-285 (2007)

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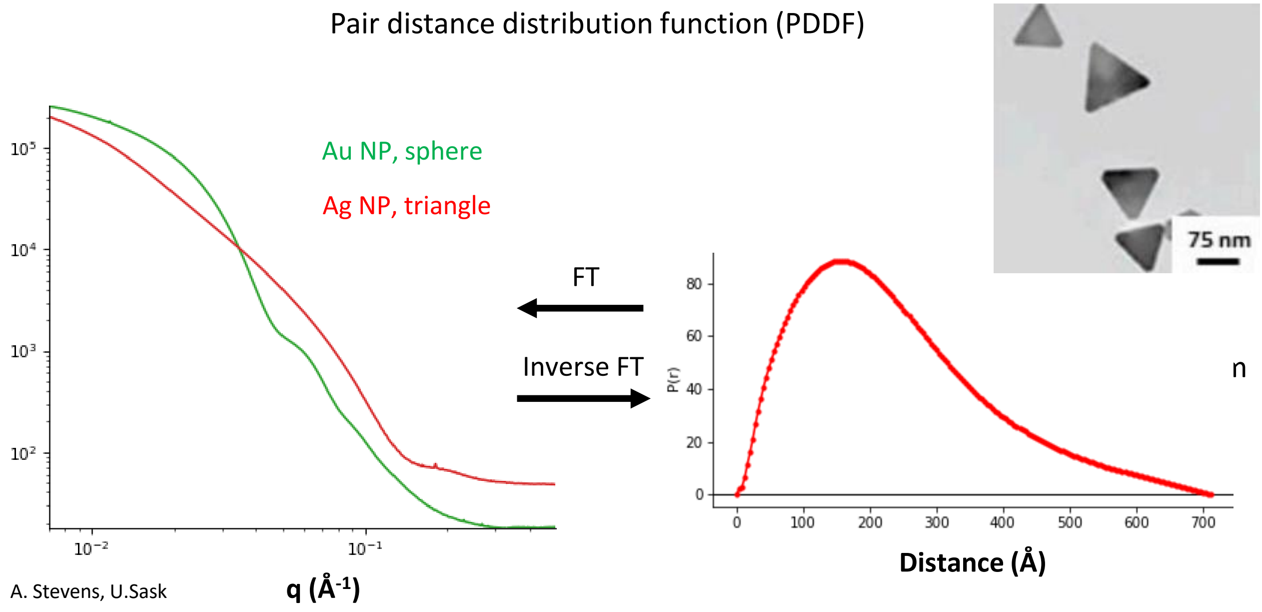
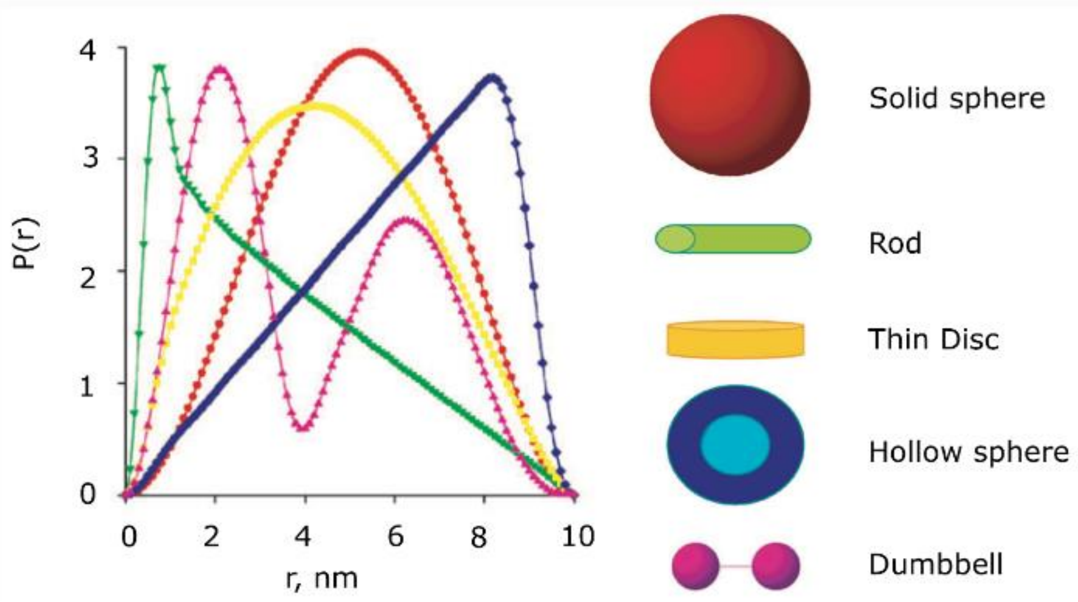
Form factor data analysis: Catalytic nanoparticles

Pair distance distribution function (PDDF)





D. Svergun, M.H.J. Koch, Rep. Prog. Phys. 66 1735 (2003)

Reciprocal space intensity

Real space distance frequency

Histogram of atom-atom distances within a single particle, weighted by respective electron densities

D_{max} – **Largest interatomic distance** in the scattering particle

Shape of the P(r) function is related to the **shape** of the particle, with low spatial resolution

q range required

$q_{min} \leq \pi / D_{max}$ (π/0.005 Å^{-1} = 62 nm)

$q_{max} \geq 2\pi / D_{max}$

D. Putnam et al. Quart. Rev. Biophys. 40, 191-285 (2007)

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Optimized Chitosan-Based Nanoemulsion Improves Luteolin Release
C. Diedrich, **I. Badea et al.,** *Pharmaceutics* **15**, 1592 (2023)

Badea Group
College of Pharmacy and Nutrition
U. Saskatchewan

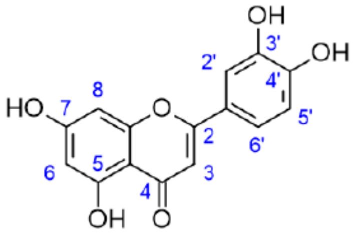
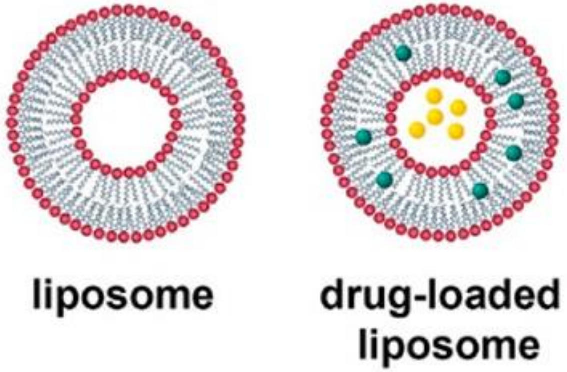


Figure 1. Luteolin structure.

Anti-oxidant, -inflammatory, -tumor, -viral
Poor absorption after oral administration, limited water solubility

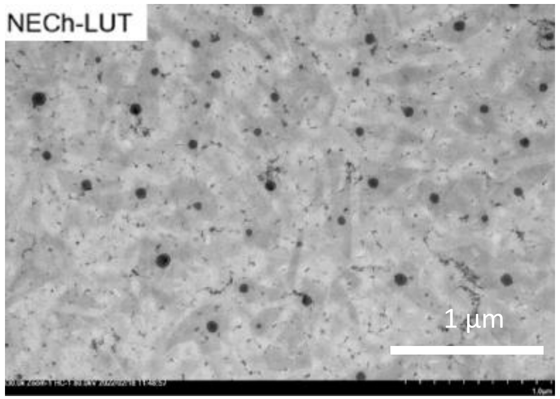
Nano-encapsulation might improve the solubility of luteolin?

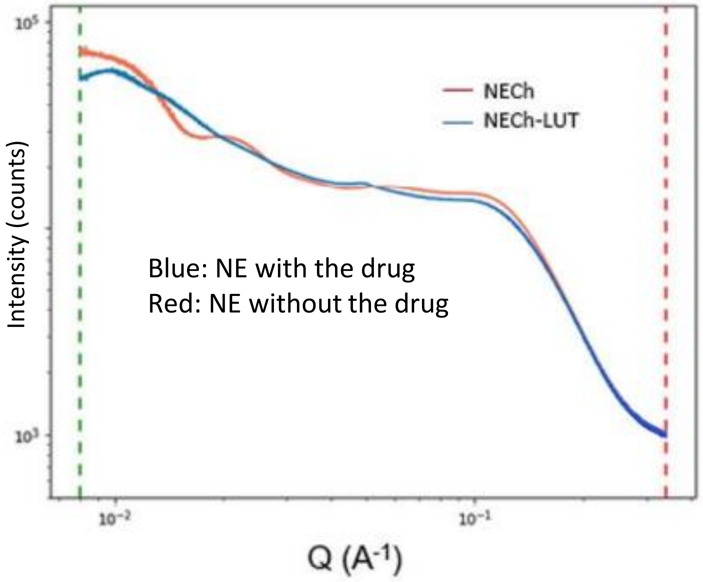
Diameter: 20 – 200 nm
Bilayer: ~5 nm



Oil phase: Luteolin, oleic acid, with ethylene glycol and Tween 20 as surfactants
Aqueous: Chitosan solubilized in 0.25% acetic acid.

Drop aqueous into oil while sonicating





The most reasonable fit was obtained modeling two spherical droplets

d = 27.0 ± 2.6 nm	Liposomes
d = 1.4 ± 0.26 nm	Tween 20 micelles

Where is luteolin?

1. **Dilute (~1 – 10 mg/mL), no interparticle interactions**
2. **Pure**
3. **Monodisperse**

Sample prep is critical !!!

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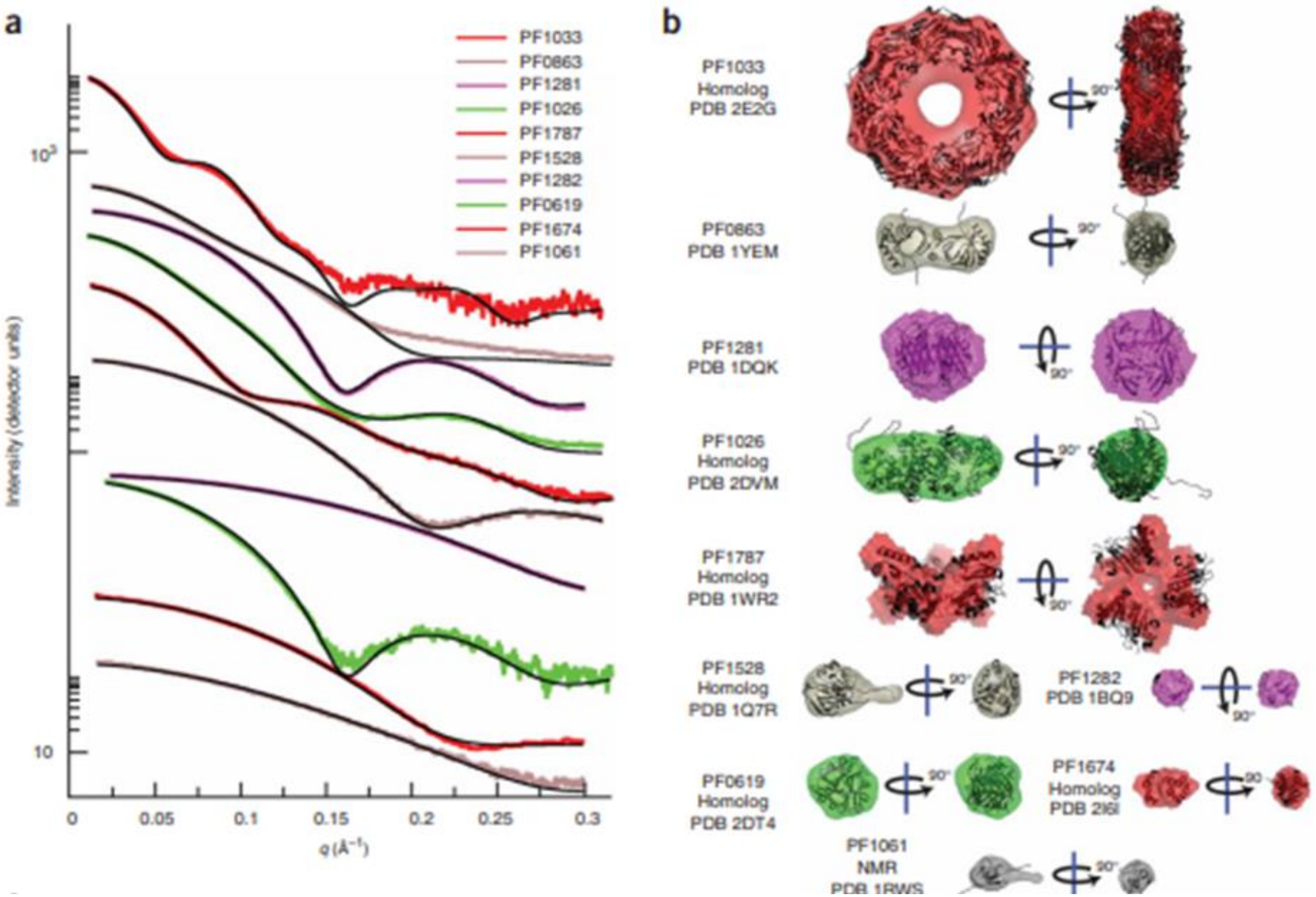
Proteins in solution

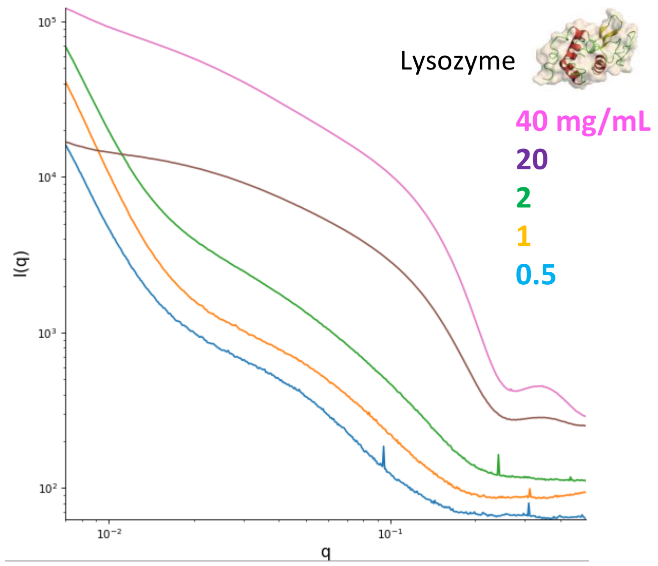
Several dedicated Bio-SAXS beamlines

Aid structure determination
- Size, molecular weight (>10%) to verify the oligomeric state
- Low resolution shape

Challenging !!
- Weakly scattering
- Radiation damage
- Limited amounts of material (~30 µL)

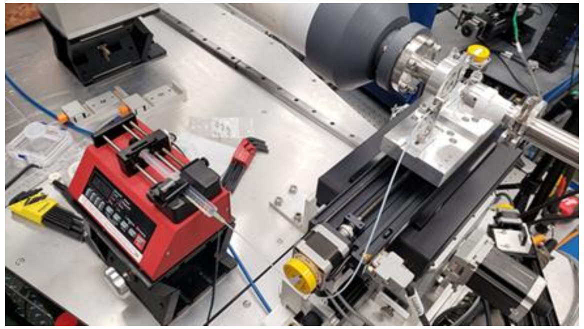


G.L. Hura et al., *Nature Methods* **6**(8), 606 (2009)



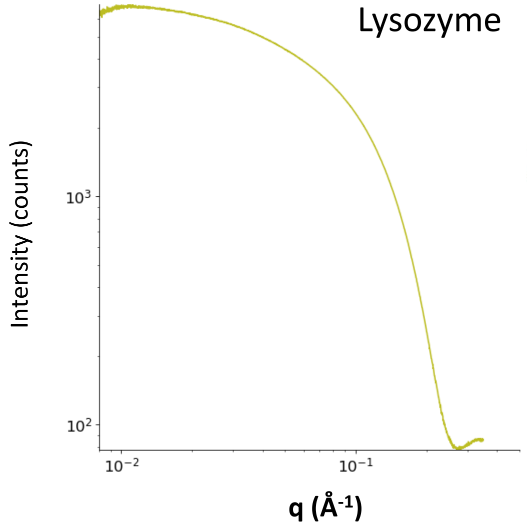
buffer

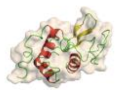
2 mg/mL



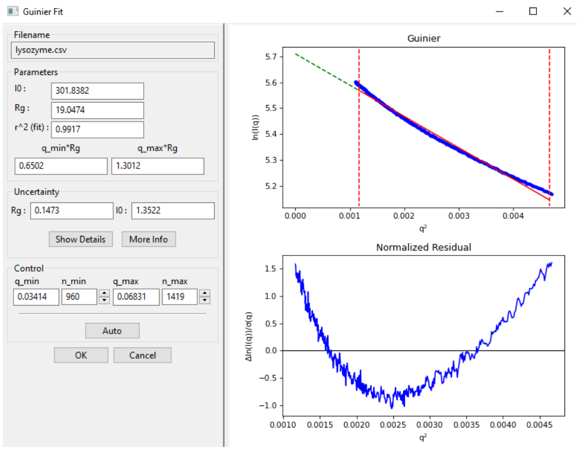
Proteins in solution

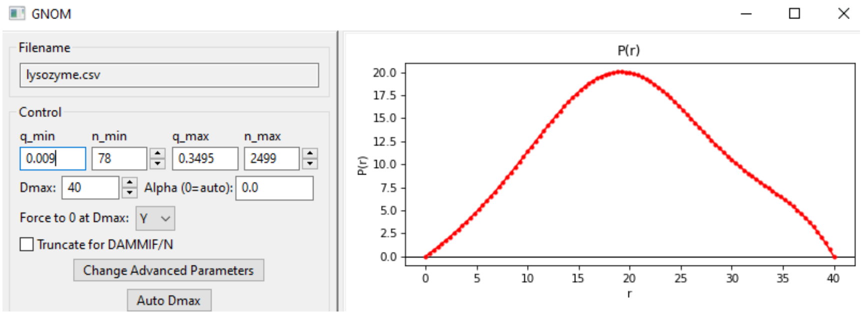
D. Svergun EMBL group example

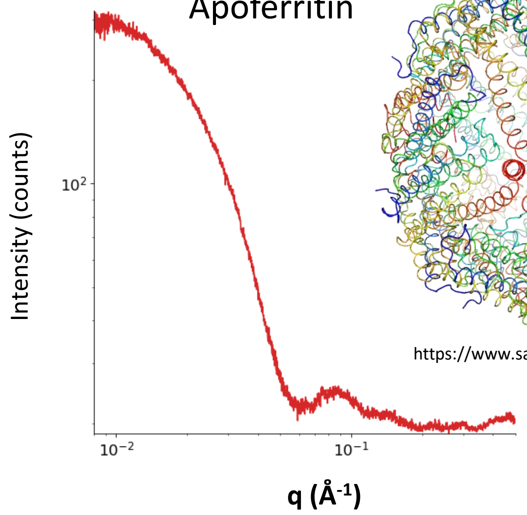


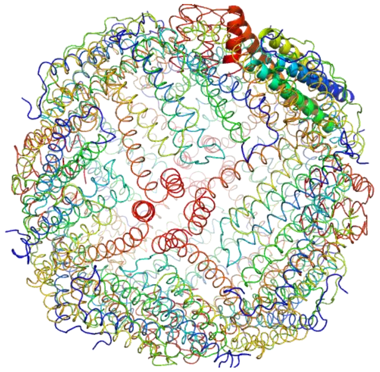


R_g: 1.9 nm



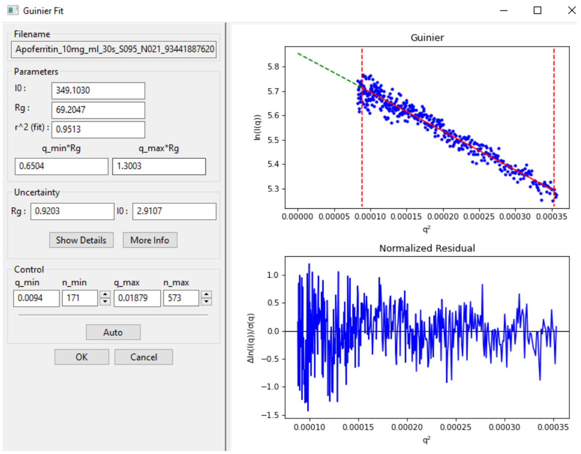


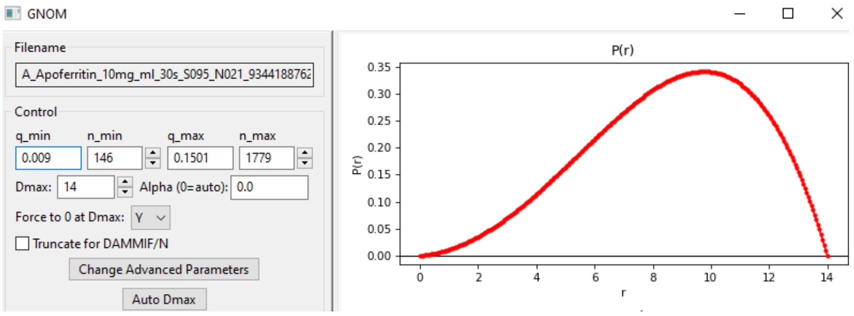




https://www.sasbdb.org/data/SASDCD8/

R_g: 6.9 nm





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Proteins in solution

https://www.sasbdb.org/

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Small Angle Scattering Biological Data Bank

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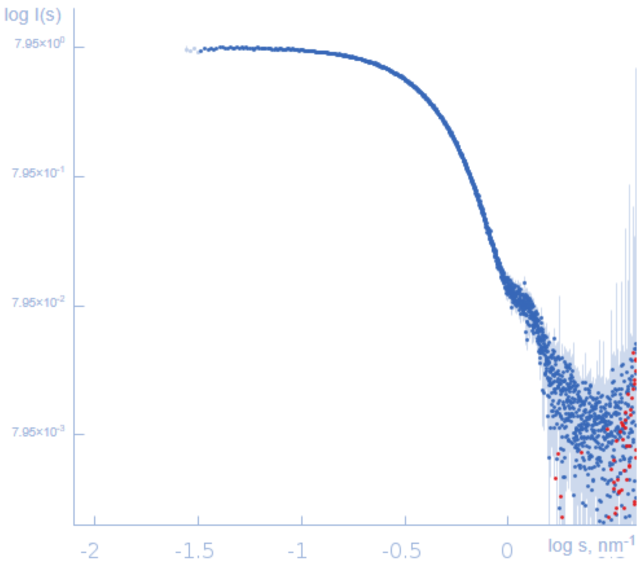
LARGE1 Processively Polymerizes Matriglycan Using Active Sites on Alternate Protomers
Joseph S, Schnicker N, **Xu Z**, Yang T, Hopkins J, Watkins M, Chakravarthy S, Davulcu O, Anderson M, Venzke D, Campbell K, *SSRN Electronic Journal* () DOI

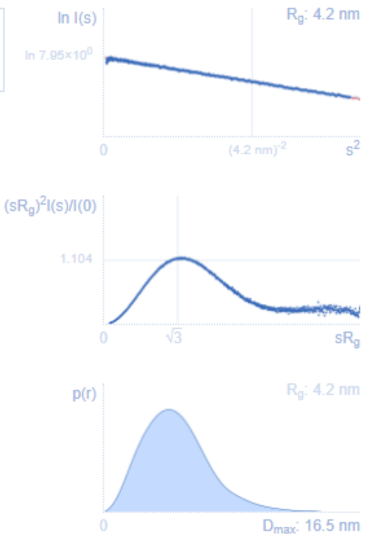
Download files ▾

SASDNH8 – Xylosyl- and glucuronyltransferase LARGE2 (LARGE2dTM) dimer

Xylosyl- and glucuronyltransferase LARGE2



$MW^{experimental}$	213	kDa
$MW^{expected}$	168	kDa



Synchrotron SAXS data from solutions of a secreted form that lacks the transmembrane domain of mouse LARGE xylosyl- and glucuronyltransferase 2 (LARGE2dTM) in buffer (20 mM HEPES pH 7.4, 150 mM NaCl) were collected on the BioCAT 18-ID-D beamline at the Advanced Photon Source (APS) (Chicago, IL, USA) using a Eiger2 XE 9M detector at a sample-detector distance of 3.67 m and at a wavelength of λ = 0.1033 nm (I(s) vs s, where s = 4πsinθ/λ, and 2θ is the scattering angle). In-line size-exclusion chromatography (SEC)-MALS-SAXS was employed. The SEC parameters were as follows: A 300 µl sample at 4 mg/ml was injected at a 0.6 ml/min flow rate onto a Superdex 200 increase 10/300 GL column (GE healthcare) at 23°C. 2500 successive 1 second frames were collected. The data were normalized to the intensity of the transmitted beam and radially averaged; the scattering of the solvent-blank was subtracted.

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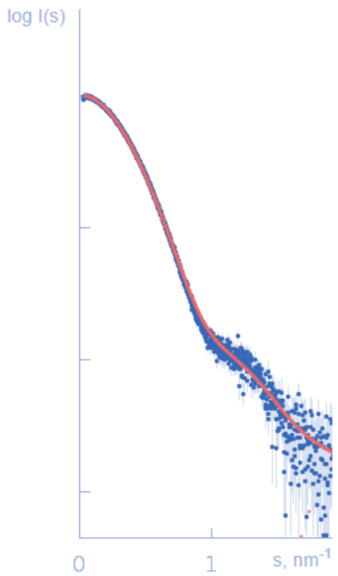
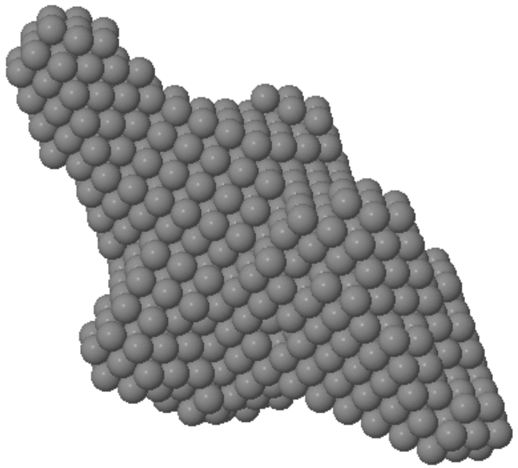
boundary conditions for bead modelling programs

Fits and models



DAMMIN model



ATSAS package
DAMMIF/DAMMIN
GASBOR

Low resolution protein structure

Summary

The CLS has a SAXS endstation available and our user community is growing!

Probe nano-scale dimensions and ordering (1 - 150 nm) with X-rays by measuring elastic scattering at small angles

Can learn about 1) packing, 2) particle size, and 3) particle shape, but generally not all 3 at the same time

Limited structural information

Complementary information, that should be supported with supplementary techniques such as microscopy

Further reading

Basic SAXS concepts

H. Schnablegger, Y. Singh, “The SAXS Guide: Getting Acquainted With the Principles”, Anton Parr, Austria. (2017)

Making a good measurement

B.R. Pauw, “Everything SAXS: small-angle scattering pattern collection and correction” J. Phys.: Condens. Matter 25 (2013) 383201

Basic data work up

J.B. Hopkins, R.E. Gillilan, S. Skou
https://bioxtas-raw.readthedocs.io/en/latest/saxs_tutorial.html

BioSAXS, data workup, introduction to structure modelling

C.D. Putnam, M. Hammel, G.L. Hura, J.A. Tainer, “X-ray solution scattering (SAXS) combined with crystallography and computation: defining accurate macromolecular structures, conformations and assemblies in solution”
Quarterly Reviews of Biophysics 40, 3 (2007), pp. 191–285.

Tutorials

Data processing
- Averaging
- Absolute intensity scale
- Data reduction

Data analysis
- Guinier plots
- Kratky plots
- PDDF
- Amphiphile phase identification

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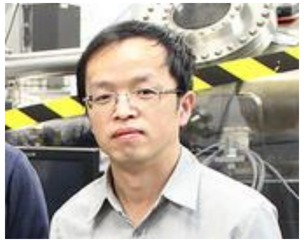

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Call for General User Proposals opens July 24, closes August 21, 2024