WP 20
« Advanced Neutron Tools for Soft and Bio Materials »

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WP 20 « ADVANCED NEUTRON TOOLS FOR SOFT AND BIO MATERIALS »

6 partners:

Participant short name	LLB CEA	HZB	JCNS	ILL	TUM	STFC
Person-months per participant (asked for EC contribution)	20	18	13	68	18	14

2 topics/ 4 tasks:

- **Platform for model biological membranes** Task 1

- **Specific sample environments for soft materials**
  - Kinetic & Dynamics experiments Task 2
  - Humidity chamber with sample changer Task 3
  - Cryogen free cryostat with sample changer Task 4

Milestones & Deliverables Task 1

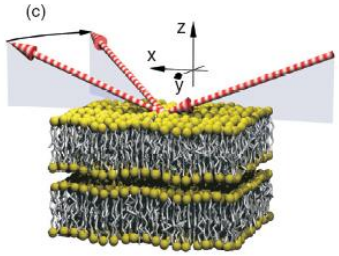
Task / Deliverable / Milestone	DETAILS	Partners in charge of task	Start month	End month	Type of deliverable
Task 20.1	**A platform for model biological membranes**	**ILL***, STFC, CEA	1	48	
M 20.1.1	Optimization of model bilayer systems including natural membrane lipids	ILL, STFC	1	36	
M 20.1.2	Set up a lipid extraction facility to extract and fractionate membranes from real cells	ILL	1	18	
D 20.1.1	Protocols for reliable reconstitution of membrane proteins	**STFC**	12	48	Report
D 20.1.2	D lipids extraction. Characterization of biomembranes with scattering and complementary techniques.	**ILL,** STFC, CEA	1	48	Report

A platform for model biological membranes Task 1

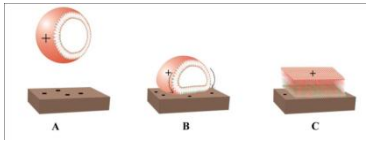
Optimization of model bilayer systems including natural membrane lipids

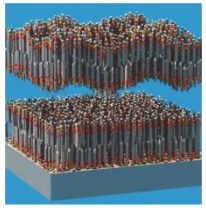
Basic Requirements of Model Bilayer for Reflectivity: large and uniform membrane

Easy to make – Vesicle fusion (or Langmuir Blodgett/Schaefer)..



But, substrate effects….

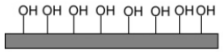


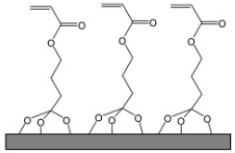


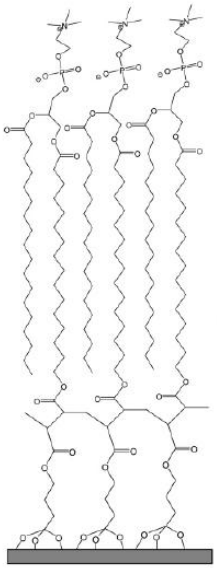
Solution - 'Floating' Bilayers..

Works – But.. only for the simplest lipids
Need to hold lower 'support' more firmly.

Solution – Grafted phospholipid on Self Assembled Monolayers





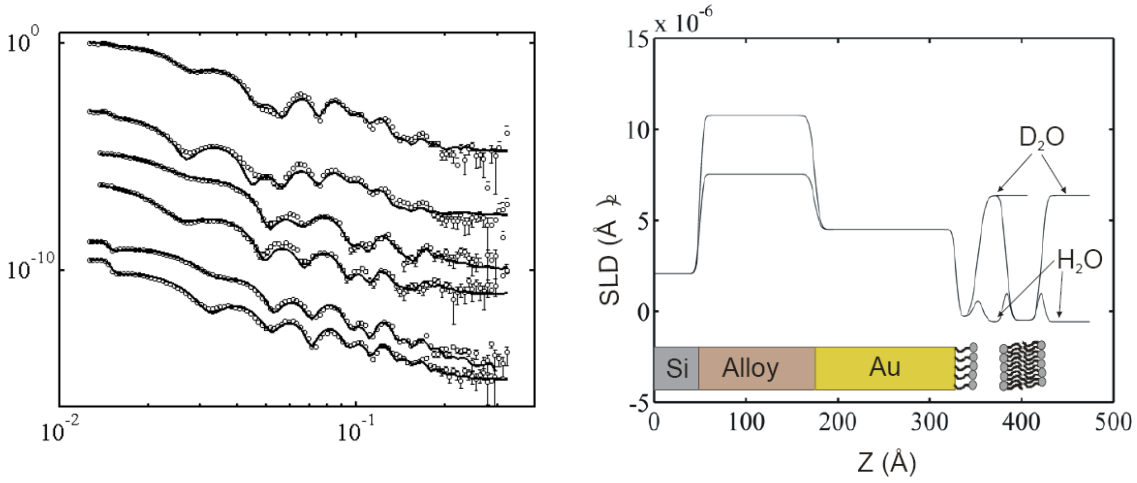


Allows fabrication of membranes from more complex mixtures.

A platform for model biological membranes Task 1

Coverage of Silane SAM never exceeds about 80%. Found that coverage of subsequent membrane usually matches the SAM coverage.

Latest developments – Gold/Thiolipids



This gives 100% coverage and subsequent improvement in membrane quality….
A.V. Hughes S.A.Holt,A.Soliakov,T.R.Charlton and J.H. Lakey et al. *Submitted to Langmuir.*

Developed novel LB technology for accurate positioning of substrates.

A platform for model biological membranes Task 1

Fully deuterated natural lipid membranes

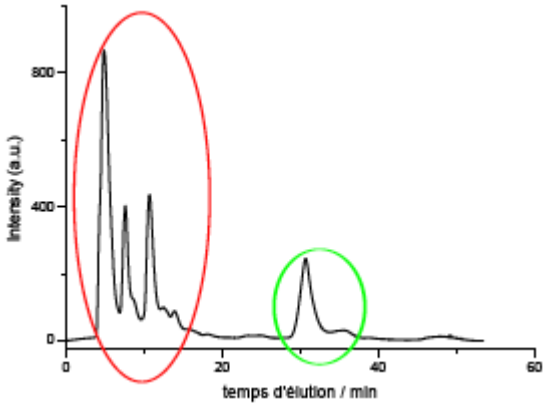
Yeast cells grown in a deuterated medium (D-lab)

Lipids extracted with Folch method Phospholipids separated by 2D TLC

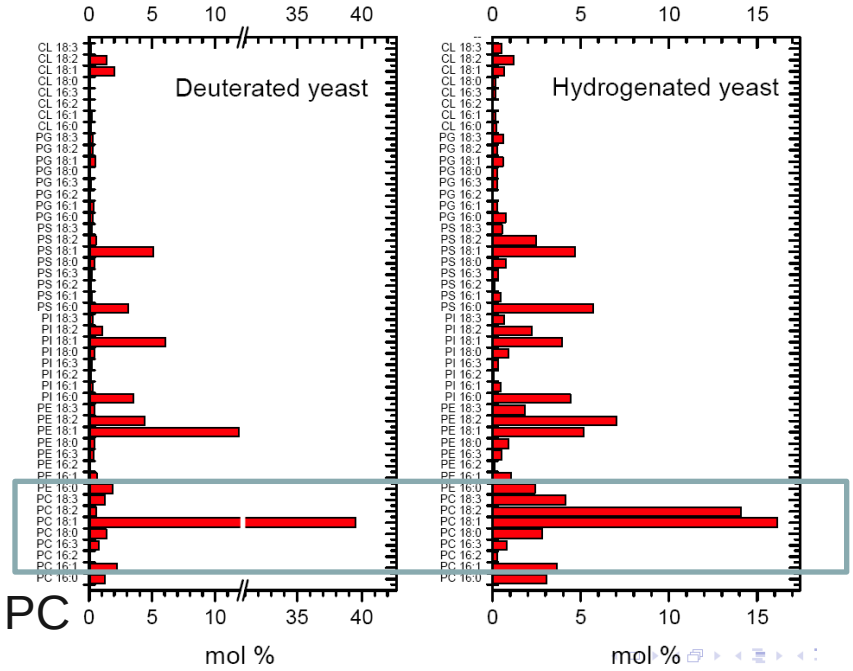
Lipids separation (HPLC):

Apolar lipids (chloroform-acetic acid)
Polar lipids (methanol)

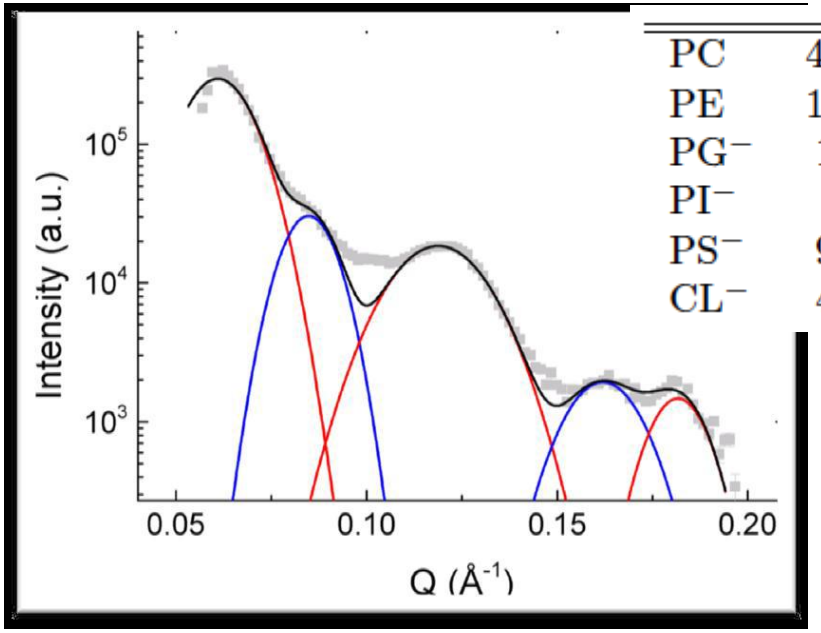
Separation of sterols from apolar



A platform for model biological membranes Task 1

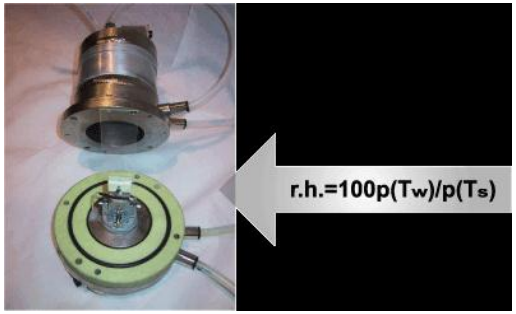


Neutron Diffraction from P. Pastoris



PC	46.8%
PE	19.9%
PG^-	1.7%
PI^-	12%
PS^-	9.8%
CL^-	4.2%

Different Humidity Ratio



	HRH		LRH	
	d_a (Å)	d_b (Å)	d_a (Å)	d_b (Å)
18°C	105 ± 2	76 ± 3	90 ± 10	72 ± 5
30°C	105 ± 2	76 ± 3	86 ± 5	73 ± 2
60°C	104 ± 2	−−	82 ± 6	74 ± 2

Y. Gerelli
G. Fragneto
A De Ghellinck .

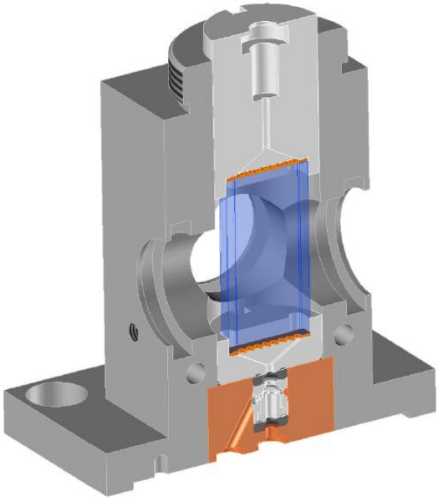
Milestones & Deliverables of Task 2

Task / Deliverable / Milestone	DETAILS	Partners	Start month	End month	Type of deliverable
Task 20.2	**Kinetic/dynamic measurements in periodic external fields**	**JCNS***,CEA,ILL	1	48	
M 20.2.1	Designs of new stop flow observation heads for SANS.	ILL	6	18	
M 20.2.2	Conception and design of MA-LS setup	JCNS,ILL,CEA	1	18	
M 20.2.3	Design an electric field cell for SANS	CEA	1	18	
M 20.2.4	Conception and design of a pressure cell for NSE	JCNS, CEA	18	36	
D 20.2.1	Tests of MA-LS prototype setup	**JCNS**, ILL,CEA		24	Report
D 20.2.2	Tests of new stop flow observation heads for SANS.	**ILL**		30	Report
D 20.2.3	Prototype of pressure cell for NSE. Tests	**JCNS**, ILL,CEA		48	Report
D 20.2.4	Prototype of EF for SANS. Tests. Design of EF for TOF.	**CEA**	18	48	Report

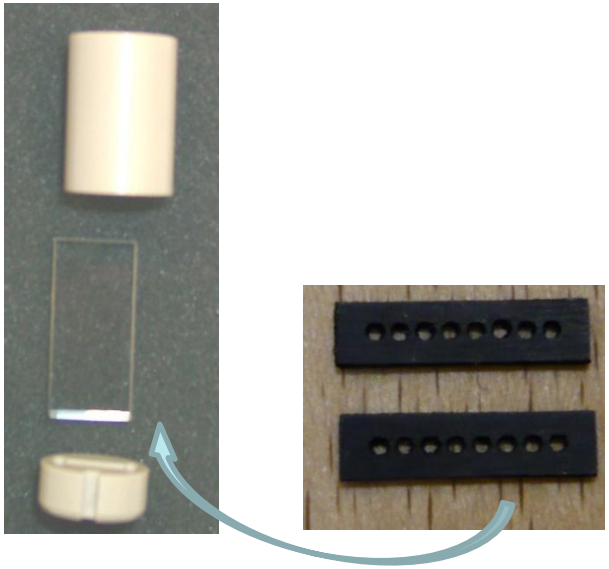
Kinetic/dynamic measurements
Task 2

New observation heads for stop flow

ILL



Hellma cell open at the 2 edges
10 x 25 x 1 mm^3 ref 690.225-QS



Cell volume 250 mL but 600 mL really needed to be sure to remove completely the old solution

… Improve the cell filling (homogeneity , duration) .

Optimize the cell geometry (not rectangular?)

Liquid flow simulations are required

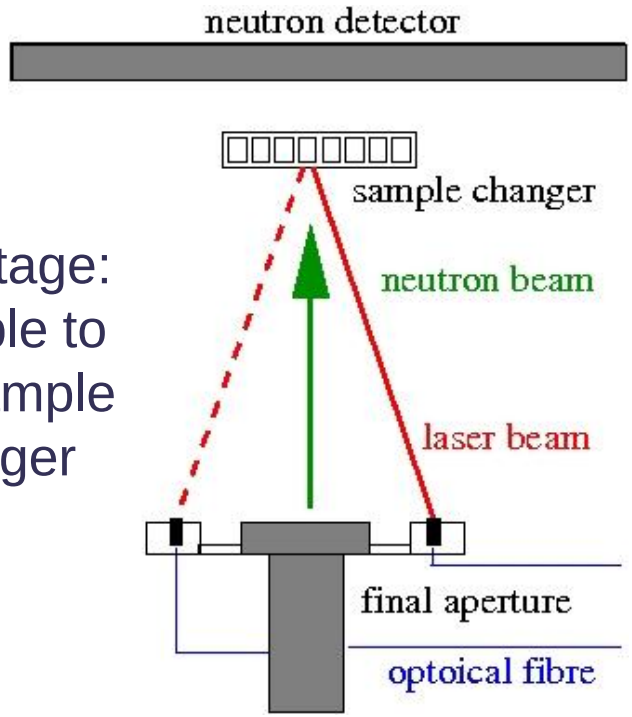
New furnace for T-jump.

Kinetic/dynamic measurements
Task 2

A combined static LS / SANS

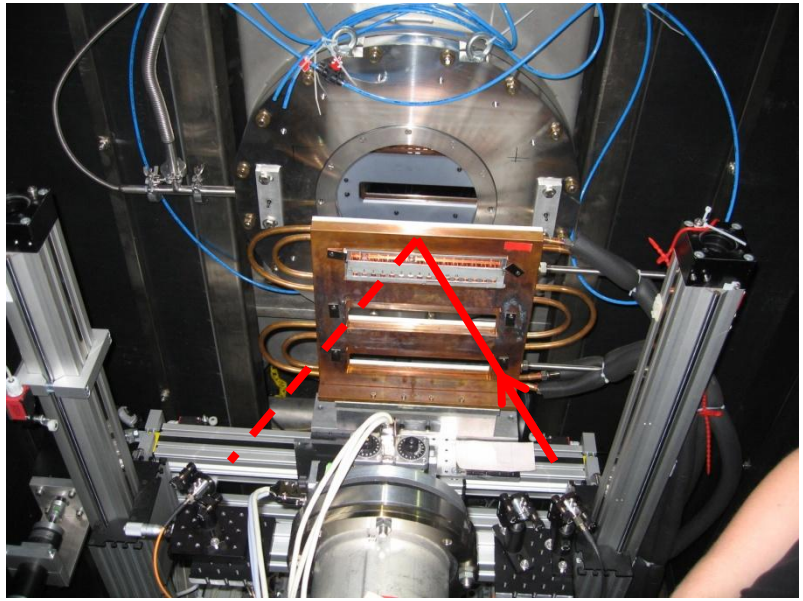
Implement **DLS …. with the flow-through cell of** the stopped flow …

LS in fiber configuration permanently fixed on the SANS collimator exit



advantage: possible to use sample changer

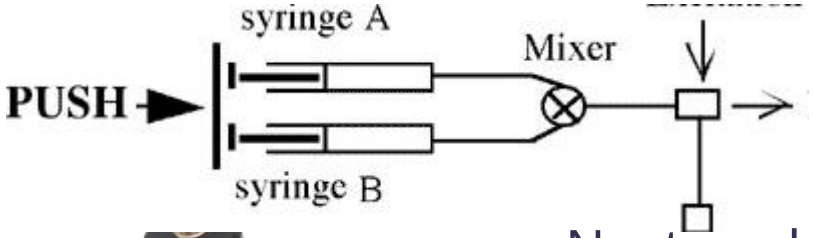
KWS2 JCNS



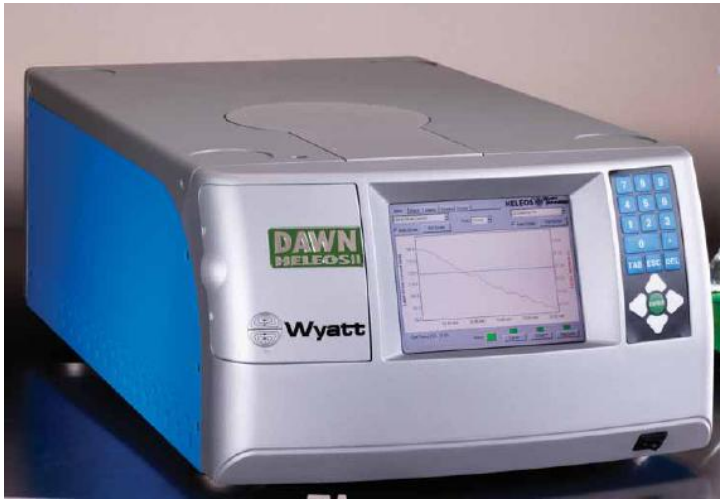
T. Schrader

… tests

Multi-angle static light scattering combined with stopped flow



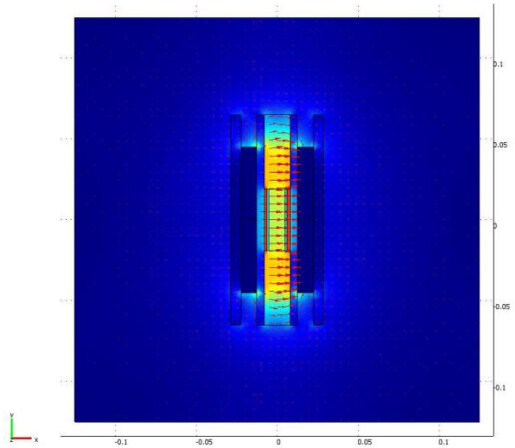
to Wyatt light scattering set up



… to install and test.

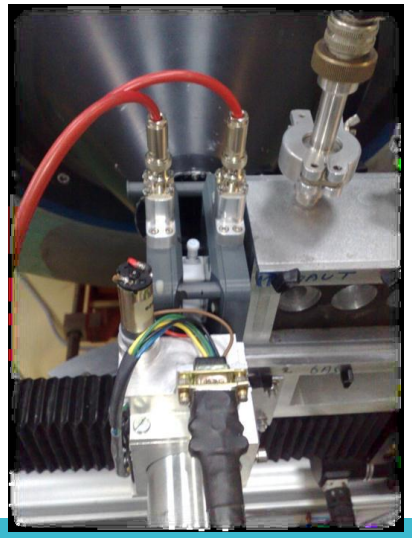
Kinetic/dynamic measurements
Task 2

Electric field cell with electrodes outside the sample



Comsol simulation

Prototype at LLB



…Tests on « reference » samples
Electric field measurement in situ ?
Thermalization (wished)
Electric field // or⊥ to the neutron beam

A. Hélary

Pressure cell for Neutron Spin Echo

Sample area: ca. 3x3 cm^2
Thickness: ca. 2mm
Pressure: 10 kbar (if possible)

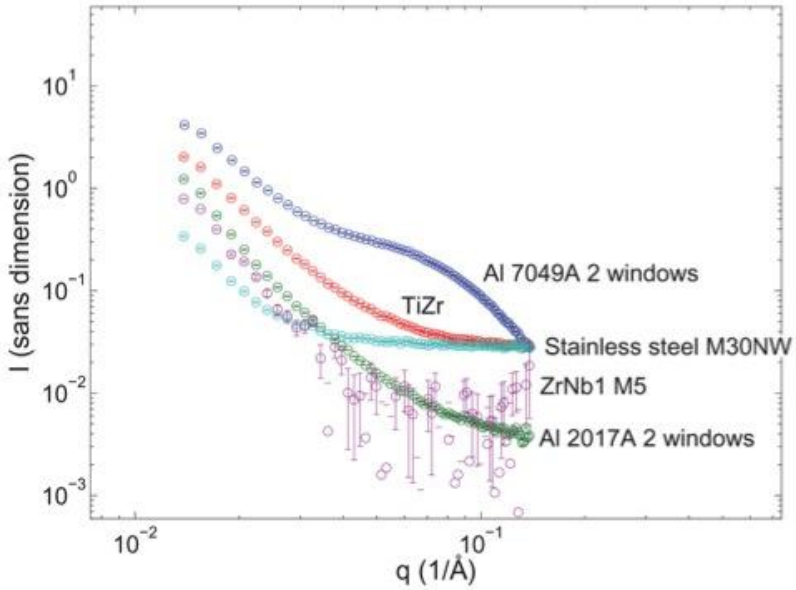
Tests of windows for Pressure cell in SANS Q Range of NSE

Window <u>thickness</u> for P_{max} ≈ 6,000 bar:

- Al 7049A: 12.2 mm
- Al 2017A: 12.2 mm
- TiZr: 8.6 mm
- Stainless steel: 10 mm

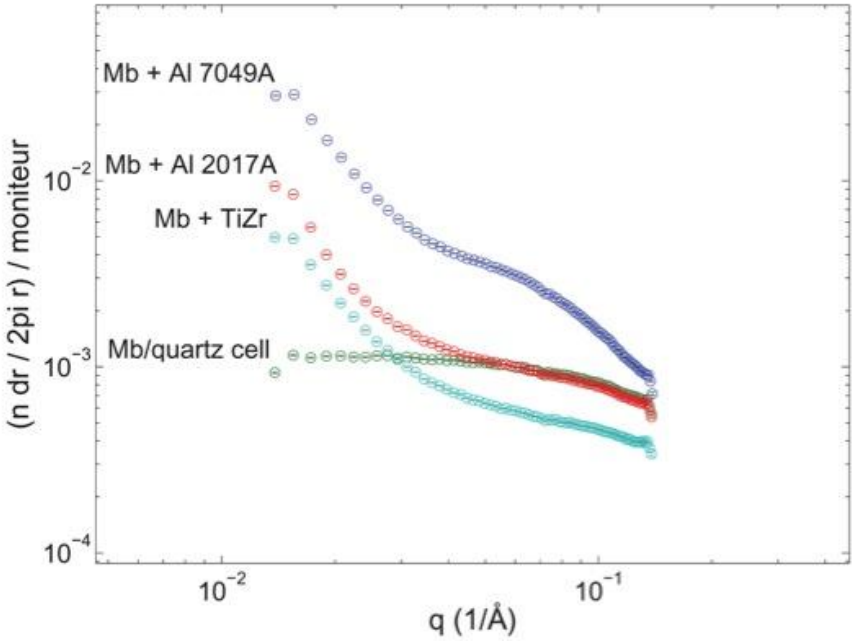
ZrNb1: 0.4 mm (thin sheet)

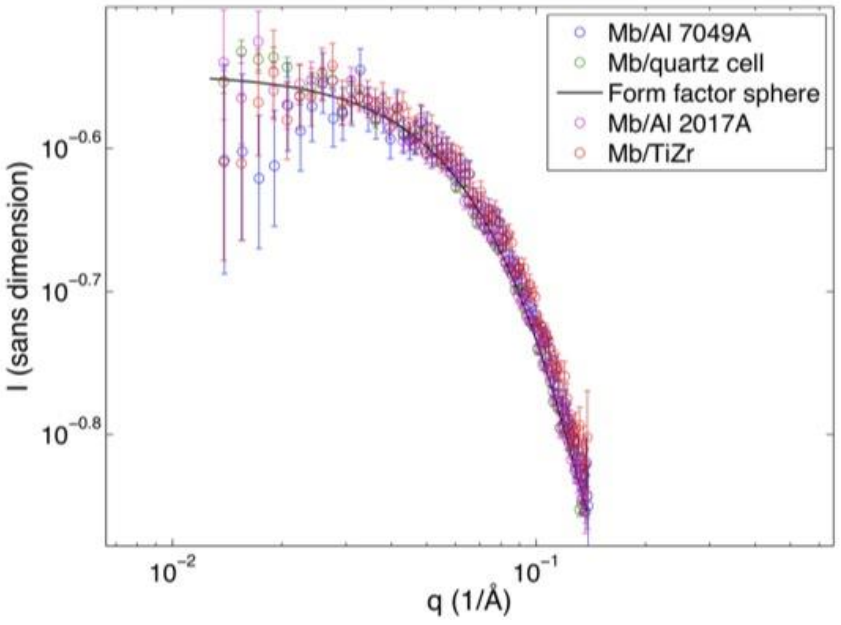
Transmission TiZr or Steel low! 0.36 /0.3 @ 6Å



Kinetic/dynamic measurements
Task 2

Windows + Myoglobine solution





All windows are OK for SANS (keep in mind the low transmission values)

… tests with NSE

What design for pressure cell of large sample area:

Flat? Collection of cylinders?

Milestones & Deliverables of Task 3

Task / Deliverable / Milestone	DETAILS	Partners in charge of task	Start month	End month	Type of deliverable
Task 20.3	**Humidity chamber**	**HZB***,ILL,JÜLICH	1	48	
M 20.3.1	Specifications of the next-generation humidity chambers. Drawings and procurement of components for humidity chambers	HZB, ILL	1	36	Drawings and Report
D 20.3.1	Assembly of the humidity chamber. Tests	**HZB**, ILL,JÜLICH	36	48	Report

Review existing systems

Summary of 22 October Meeting

	Humidity ceiling (at 25 C)	Humidity Stability	Automation for RH change	Equilibration time (after RH change)	Contrast variation (H_20/D_20)
Gas flow	~95%	~1% (to be tested)	MFCs	minutes	Bubble through mix
Saturated salt	98% (discrete steps)	Weak temperature dependence	Syringe pumps (Sat. and distilled)	hours	Change of sample/ flow into reservoir
Temperature controlled	≤100%	~1% (to be tested)	Peltier/water bath temp	hours	Flow liquid into reservoir
Fixed humid	Same as salt	No regulation	X	X	Change of sample
Bulk water	Saturated	Stable	X	X	Flow into bulk volume

Humidity chamber Task 3

Specifications of

New Humidity Cell

Consensus

- Double walled aluminum cylinder
- Isolation of base from walls (Teflon or ceramic)
- High humidity necessary
- Fast equilibration and long stability necessary

To test

- Gas flow or water troughs
- Addition of circulation fan
- Sensor accuracy and reproducibility
- Finding best 'characterization sample'
- Several samples in one can/several identical cans

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HZB Helmholtz Zentrum Berlin

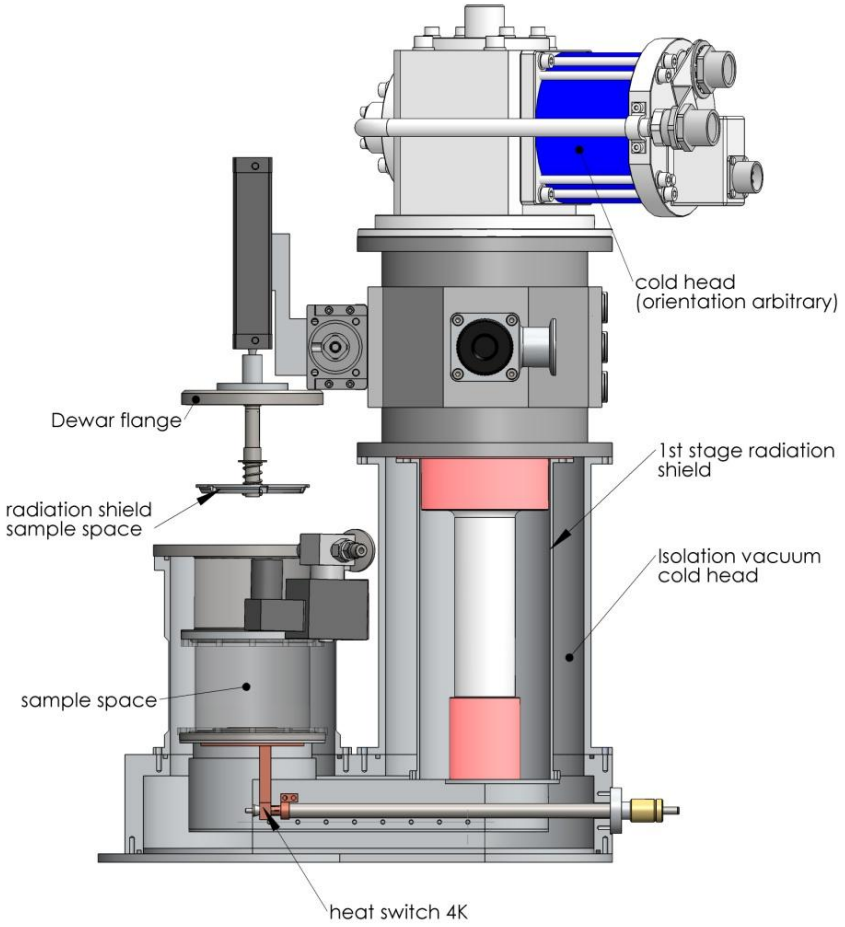
Task 3: Humidity Chamber

Milestones & Deliverables of Task 4

Task / Deliverable / Milestone	DETAILS	Partners in charge of task	Start month	End month	Type of deliverable
Task 20.4	**Cryogen-free cryostat with sample changer**	ILL,**TUM***,STFC **change**	1	48	
M.20.4.1	Design and performance estimations of cryostats with sample changer.	ILL,TUM	1	36	Report
D 20.4.1	Drawings of the cryostat.	ILL,TUM,STFC		48	Report

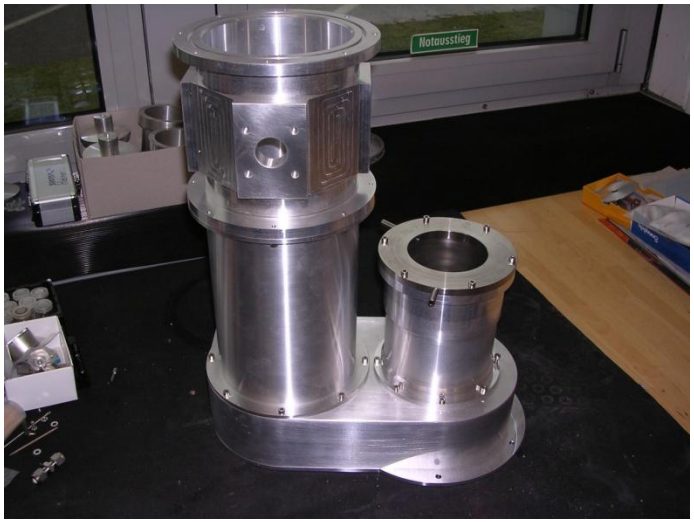
Cryogen-free cryostat with sample changer Task 4

- **Compact cryostat:**
  - Separate sample space and cold head isolation vacuum
  - Minimized cold mass
  - Remote controlled reload
  - Standardized sample holder
  - Pin connection for thermal link and thermometry
  - Sample in exchange gas via sample container

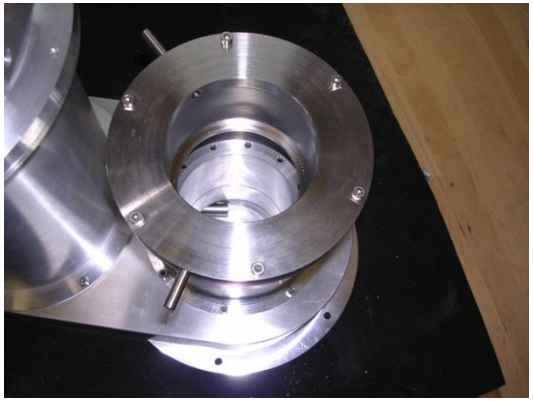


Cryogen-free cryostat with sample changer Task 4

Dewar

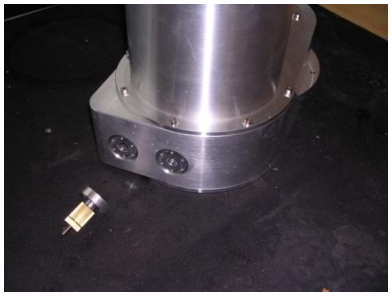


Radiation Shield



Nested links

Heat switch



- **…. to be solved**
  - Efficiency of heat switch
    → Cool down time
    → Temperature
  - Thermal connection of sample
    → Pin connection
    → Standardized sample cans

J. Peters