

6 partners:

Participant short name	LLB (CEA)	HZB	JCNS	ILL	TUM	STFC

4 tasks:

- Platform for model biological membranes Task 1
- Kinetic & Dynamics experiments Task 2
- Humidity chamber Task 3
- Cryogen free cryostat with sample changer Task 4

Last meeting:

May 15 - 16 2014 at ILL



nmi3



Task 1

A platform for model biological membranes

Optimization of model bilayer systems including natural membrane lipids studied by neutron reflectometry

ILL, STFC

Up-date on staff resources

- ♣ A post-doc hired at ISIS in 2013
- ♣ Yuri Gerelli (post-doc at ILL) - end NMI3-II contract Dec 2013 (model membrane systems).
- ♣ Alexis de Ghellinck - PhD Dec 2013 (lipid extraction -not funded by NMI3-II but contributed to project).
- ♣ PhD studentship awarded by ILL co-financed by ESS to continue work on lipid extraction towards deuterated lipid platform setting up. Start Oct 2014. ILL part financed by NMI3-II.

+ Means of PSCM @ ILL

- Lipid Extraction/ characterisation ILL
- Model Membrane Preparation by LB-LS/tensiometer*

+ Means of STFC Lab.



nmia3



Task 1

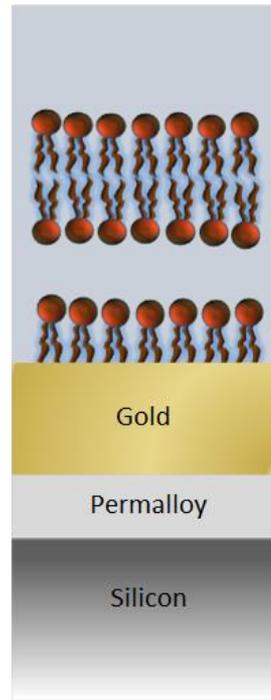
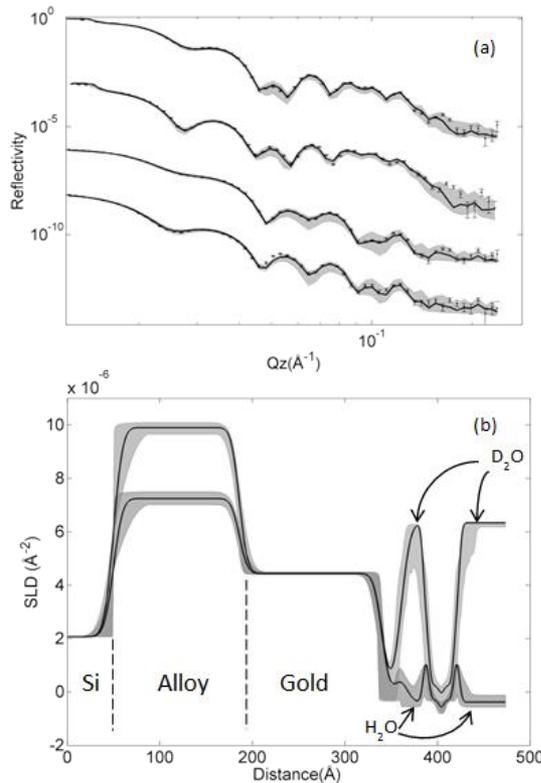
A platform for model biological membranes

Optimization of model bilayer systems including natural membrane lipids studied by neutron reflectometry

ILL, STFC

New floating membranes : Bilayers supported on thiolipid on gold

ISIS

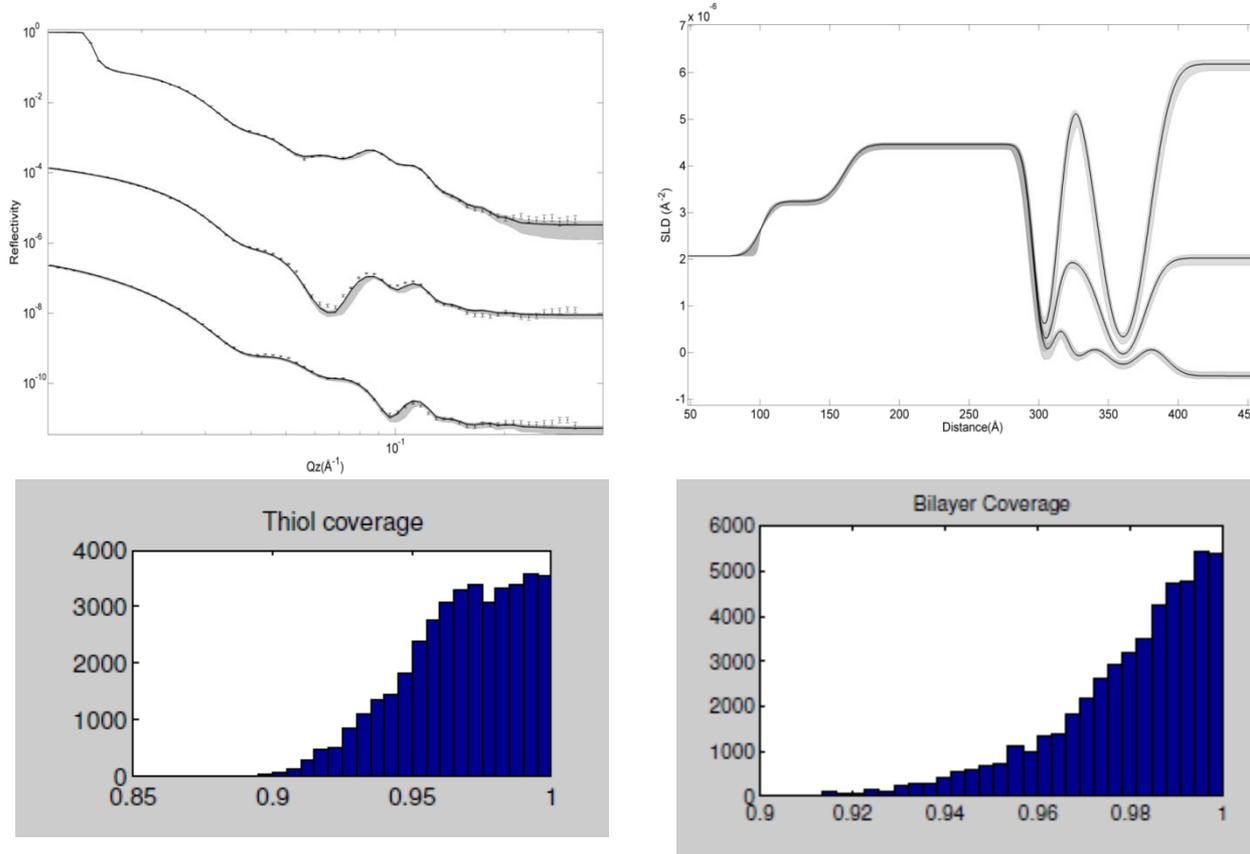


This system is giving us 100% coverage bilayers.

Now use of **magnetic underlayers** and **Polarised Neutrons** to give additional contrasts.

(ANSTO, NIST)

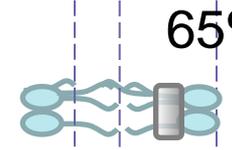
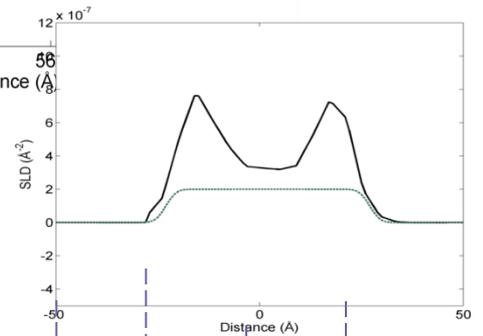
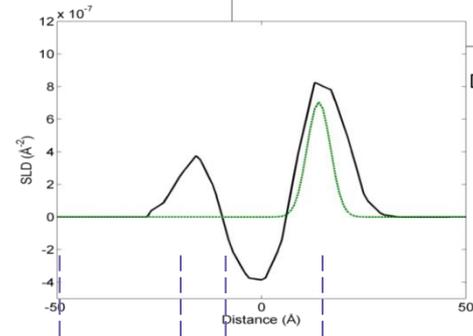
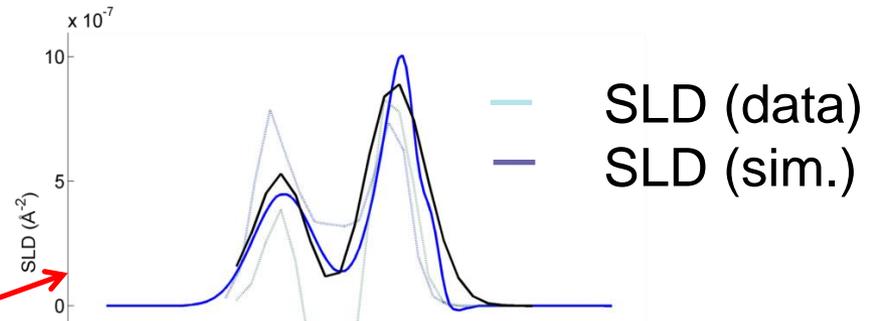
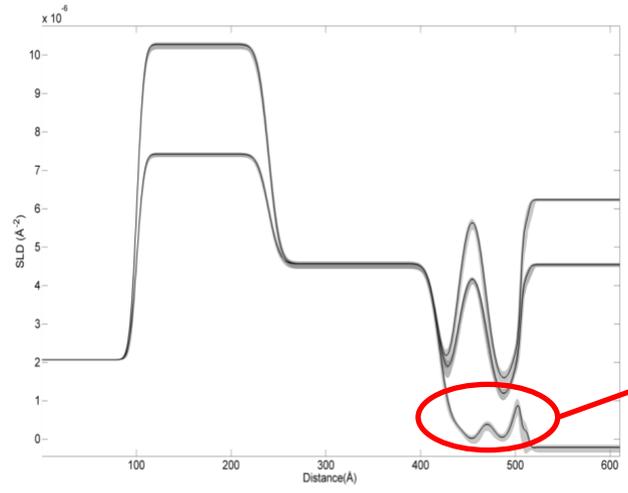
Data Analysis Development of Bayesian analysis codes for model fitting...



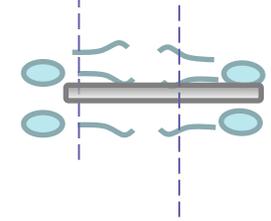
This gives robust methods for parameter (and uncertainty) estimation for 'traditional' scattering models. This is in a beta version soon ready for release...

Data Analysis

... combined with molecular dynamics



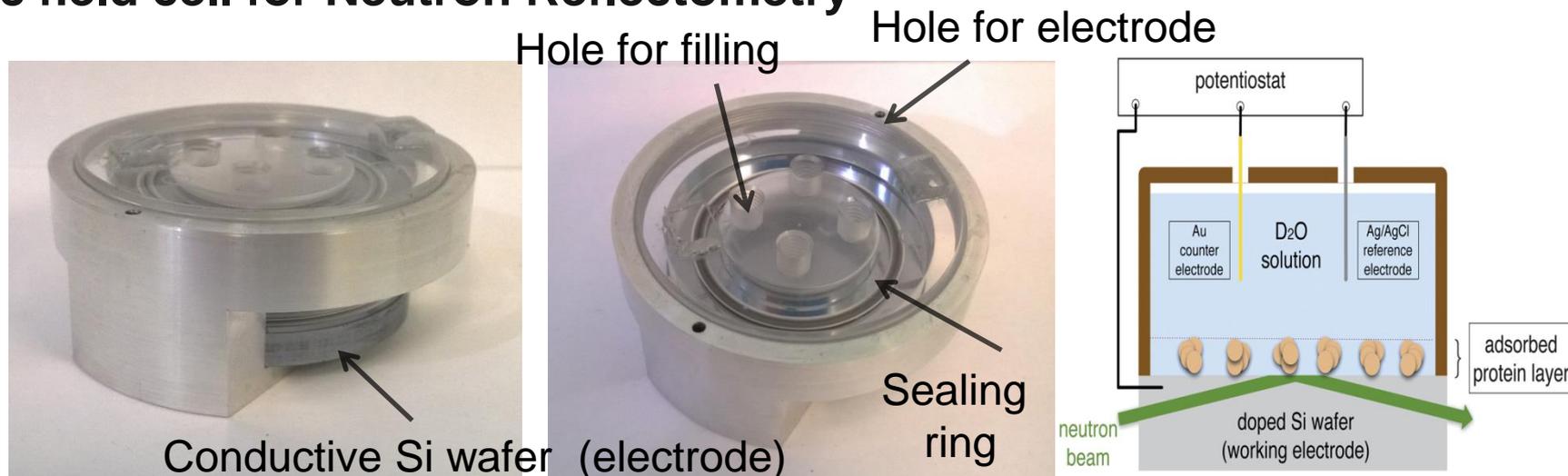
65% parallel



35% trans membrane

Electric field cell for Neutron Reflectometry

LLB



A. Koutsioubas et al. *Soft Matter*, 2012, 8, 2638

Design a new liquid liquid cell for Neutron Reflectometry

ILL

- **Optimised** neutron path **length** through liquid
- Smaller sample volume, **reduced meniscus**
- **Windows made from quartz glass, easy cleaning, etc.**



nmia3



Task 1

A platform for model biological membranes

Publications Lipid extraction including D lipids

- “Production and analysis of perdeuterated phospholipids from *Pichia pastoris* cells”. A.de Ghellinck et al.
2014 PLoS ONE 9(4): e92999. **Deliverable D20.2 M18**
- *(submitted) “Multi-lamellar organization of fully deuterated lipid extracts of yeast membranes: A neutron diffraction study”. Y. Gerelli et al..*

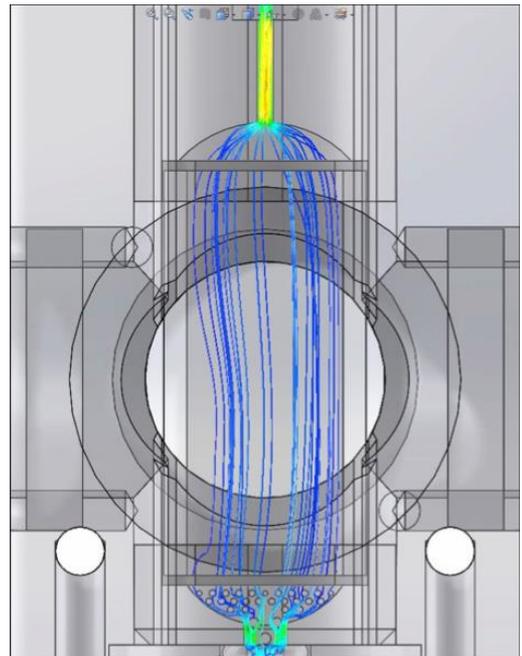
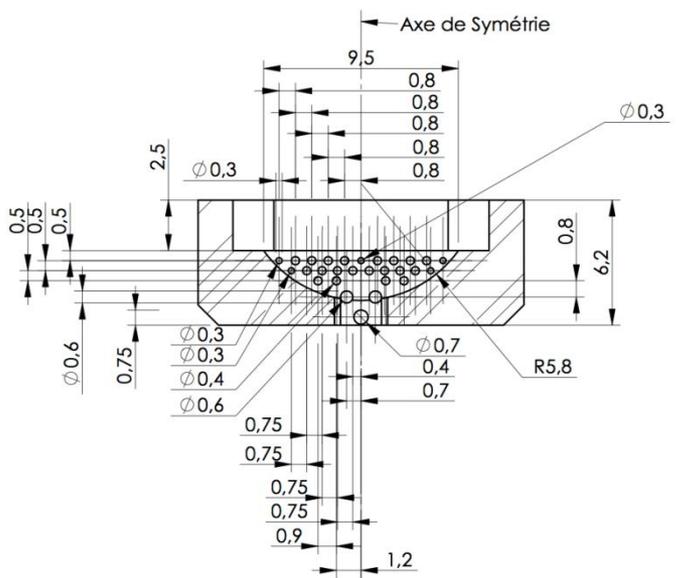
Membranes via Neutron reflectometry

- “Investigation of DPPC bilayers internal structure with specular neutron reflectometry” M. Belicka et al.
2014 J. of Colloid and Interface Sci. , 1-20
- “Lipid Exchange and Flip Flop in solid supported bilayers” Y. Gerelli et al.
2013 Langmuir 29
- *(in preparation) “Incorporation of amphotericin B in *P. Pastoris* membrane revealed by neutron reflectometry”. A. de Ghellinck, et al.*
- “High coverage fluid-phase floating lipid bilayers supported by ω -thiolipid self-assembled monolayers” A.V. Hughes et al.
2014 J. R. Soc. Interface 11(98):20140245

New observation heads for Stop Flow ILL

- Reduce wasted sample with improved mixing process
- Improve temperature stability, reuse existing syringes (very costly)

Design and simulation

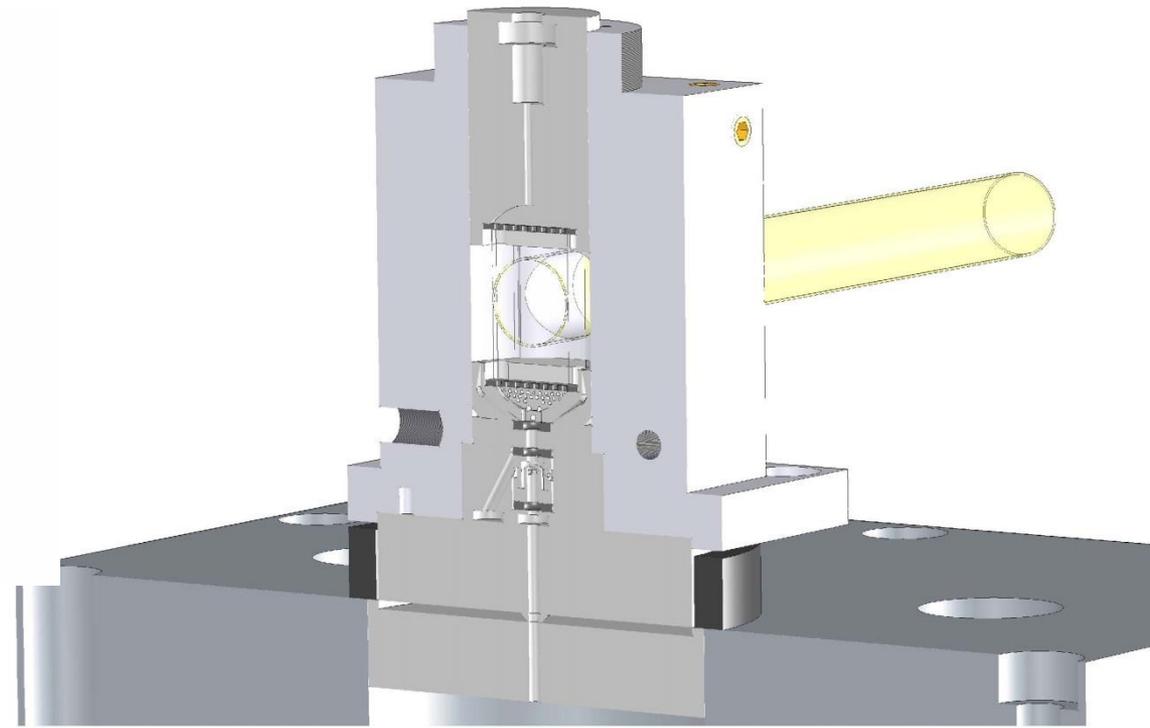
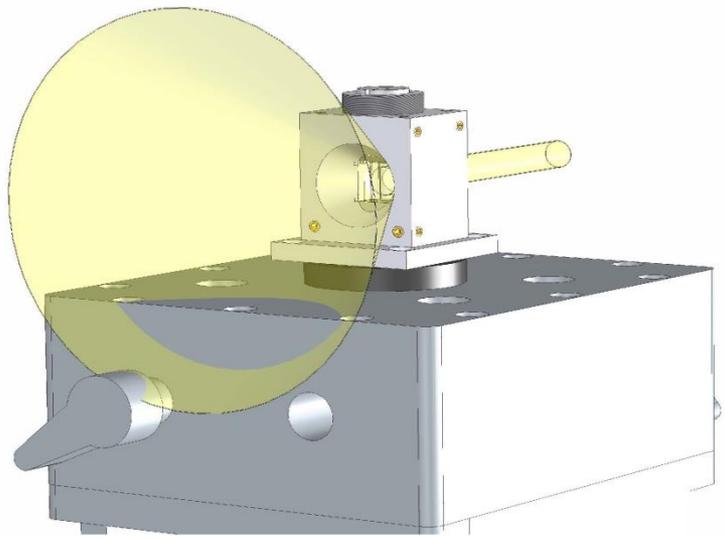


Damping grid designed at ILL, built at ISIS, and successfully tested at ILL



Design of a new temperature-controlled chamber (almost finished)

- Improve T stability with fluid circulating inside the head (0.1 K)
- Allow T jump.

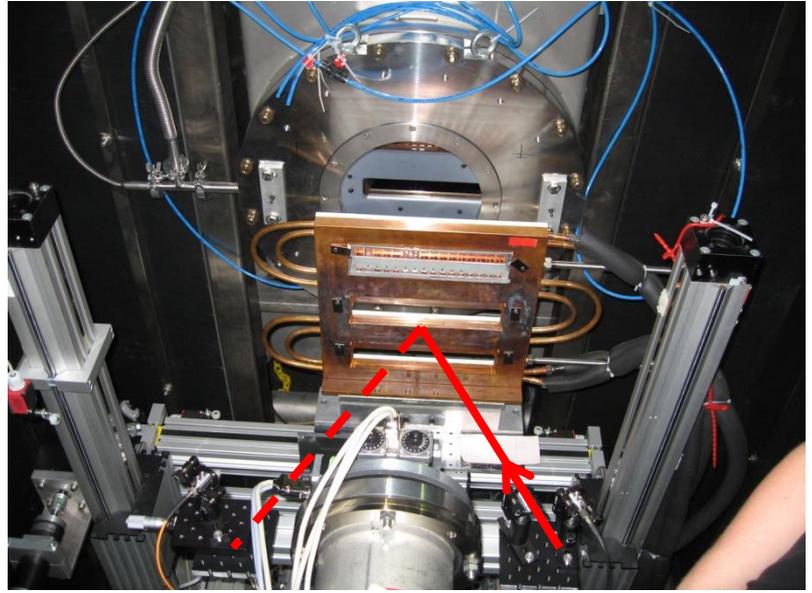
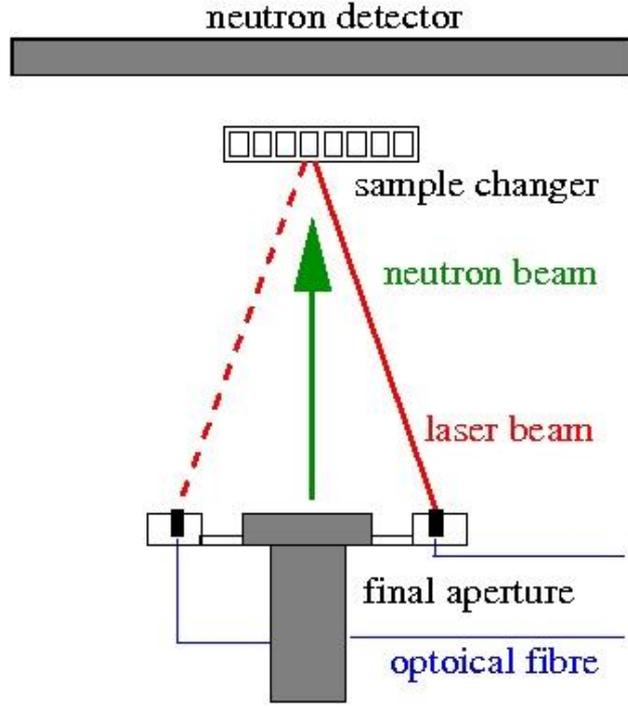


A combined static LS DLS and SANS JCNS,CEA,ILL

LS in fiber configuration permanently fixed

- 1st location : on the SANS collimator exit (JCNS)

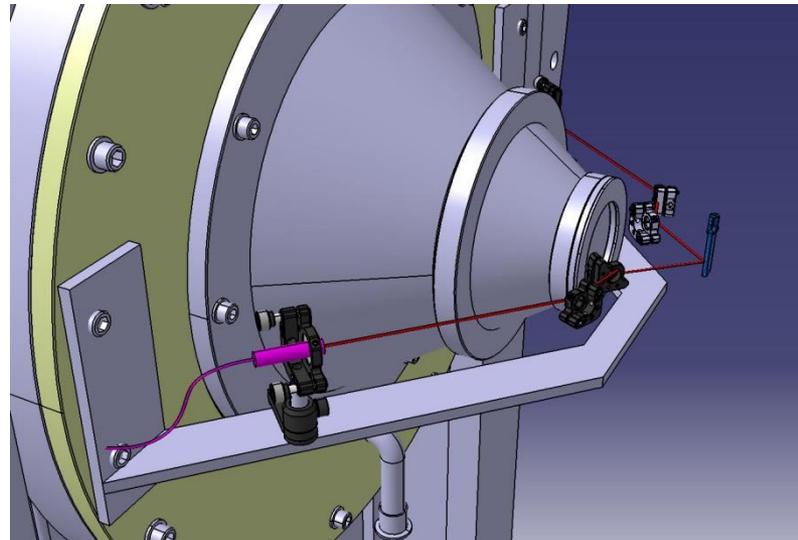
Advantage:
possible to
use sample
changer



Deliverable D20.6 M18

A combined static LS DLS and SANS JCNS,CEA,ILL

2nd location: on the detector tube nose



LS with stop flow at JCNS on going

Spreading LS /DLS /SANS ... Tests **Deliverable D20.9 M24 postponed**
JCNS experiments at the end of September at ILL

Electric field cell with electrodes outside the sample

LLB

Electric field: from 0.04 to 4 kV/cm

Temperature: from 20 to 60 °C

Prototype for classical Hellma cell at room T° ✓

Measurement of effective EF ✓ (probe at the sample location)

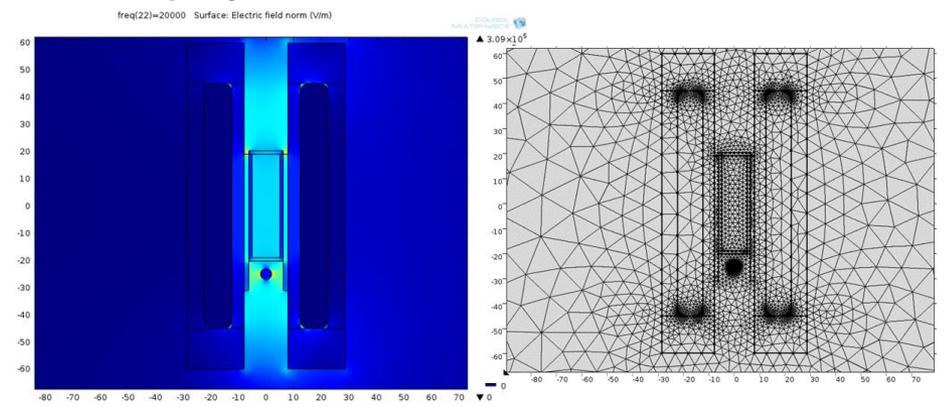
≠ solvents and materials

Fluid	Permittivity ϵ_r	Electric field (kV/cm) at 2kV 10Hz 20° C 2.5cm
Air	1.0	3.07E-1
Toluene	2.3	2.45E-1
Ethanol	24.3	2.36E-2
DMSO	46.7	5.32E-3
Distilled water	78.6	2.72E-3



Probe Kaptéos Cie

Comparison Tests / Simulations ComSol Multiphysics (visit of LLB at HZB)



Low EF

-> Get rid of materials around the cell

Design of new closed EF cell+Thermalization:

- Bath to thermalized the electrodes

bad thermalization



- Flow of thermostated gaz inside the EF cell.
on going ...

Pressure cell for Neutron Spin Echo

JCMS, ILL, LLB

For NSE: Sample area: 3x3 cm²
 Pressure? as high as possible ... 3kbar?
For SANS: Sample area: 1x1 cm²
 Pressure: 10kbar ?

Choice of materials : Sapphire or alloys windows?

Sapphire : no scattering, transparent but fragile...

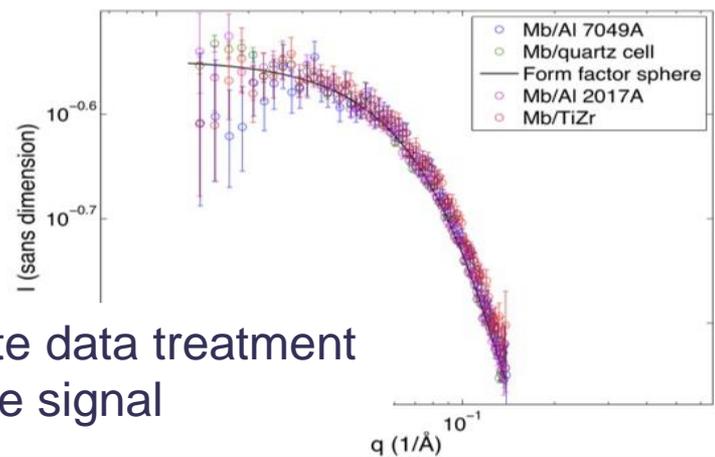
Alloys : spurious scattering but high pressure up 10kbar?

Nb : 2.7 2017A : 4.5 7049AT: 6.3kbar (LLB)

Tests of alloy windows

Al 4G or 7049, TiZr, steel, Nb, NbZr...

Q Range of NSE



Appropriate data treatment
recover the signal

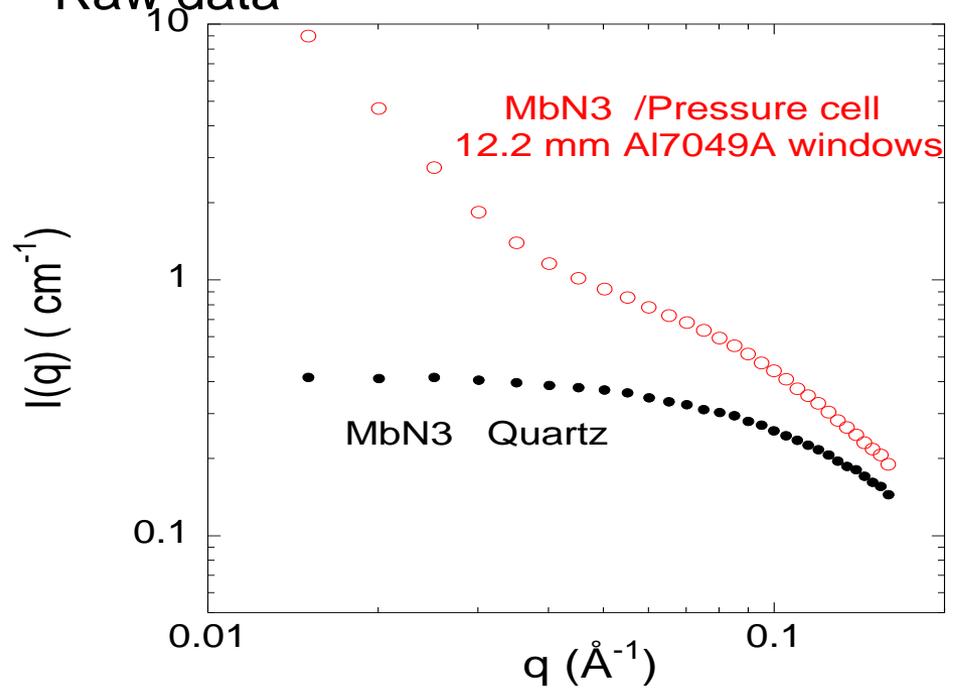
Tests at LLB

Pressure on solution of Myoglobin N3 $c=2\text{mg/ml}$

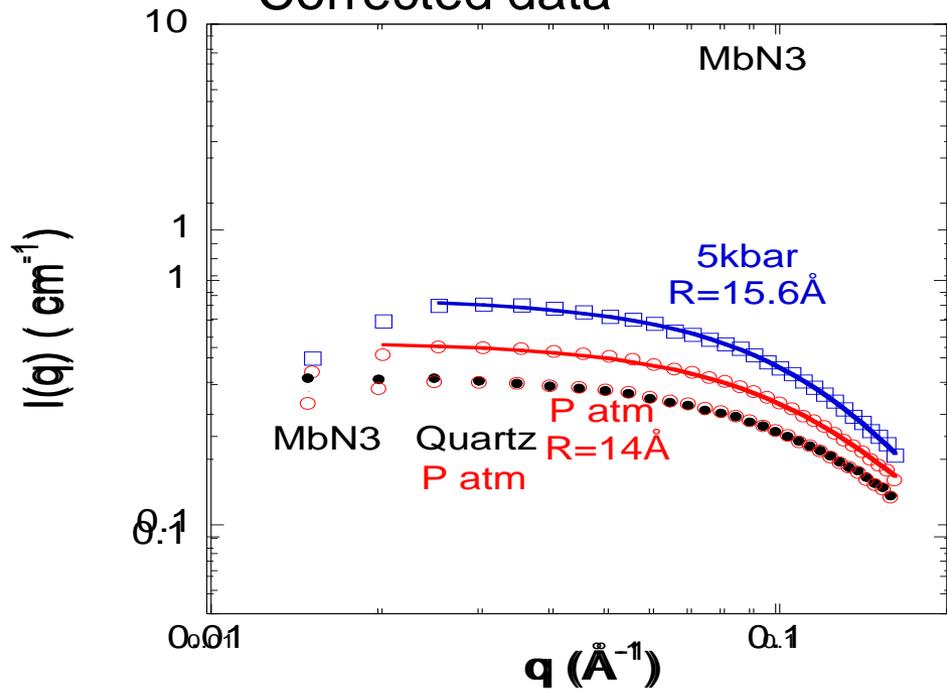
12.2mm thick Al7049A windows

6\AA 2m access limited to $\pm 10^\circ$

Raw data



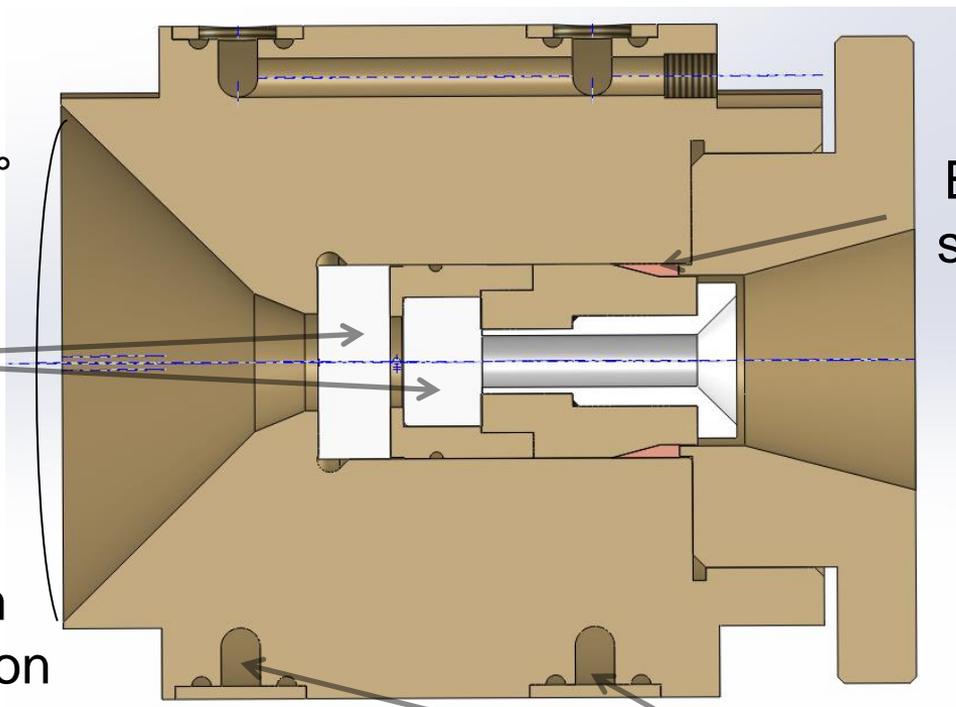
Corrected data



Design a new P cell for SANS (and NSE) up to 5-7kbar

ILL, LLB

- Large access $\pm 22.5^\circ$
-> large q
- Just cylinders
- Sapphire windows
- Alloy windows
-> Wavelength range from 5.5\AA (Multiple diffraction on polycrystalline materials) to 10\AA (multiple scattering from grain boundaries)



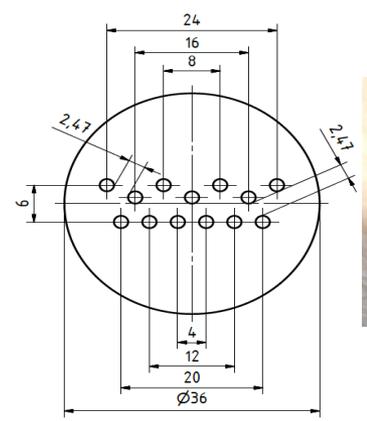
Bridgeman sealing ring

Loops for controlling temperature

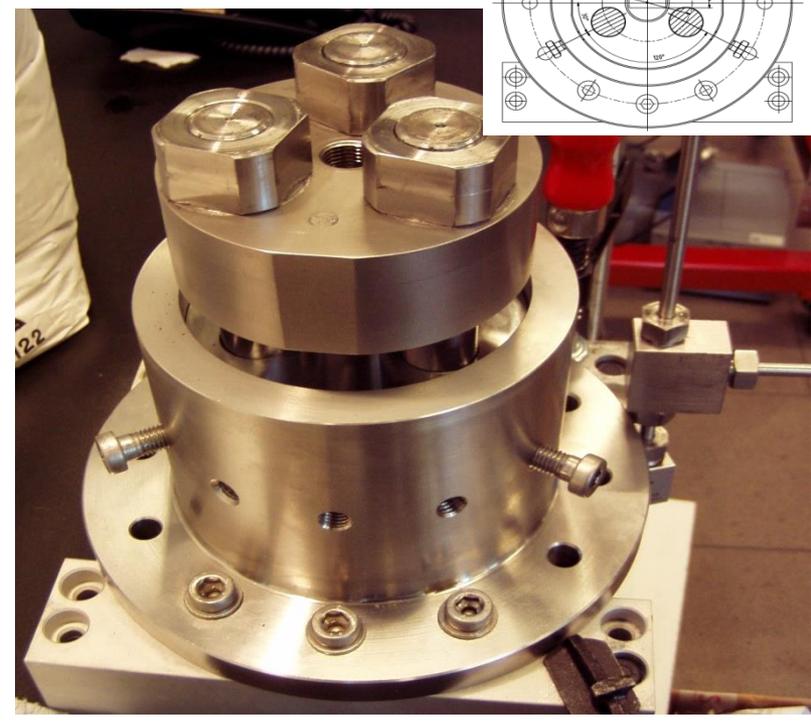
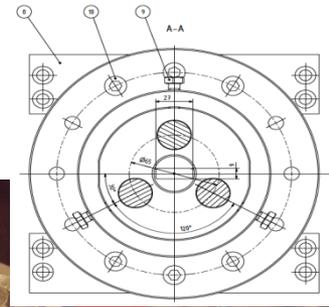
1st prototype in CuBe for NSE... 1 postdoc at LLB 11 2014

Pressure cell for NSE JCNS

Prototype Cell #1



TiZr



- Testing pressure 3.5 kbar
- operation at 2.4 kbar

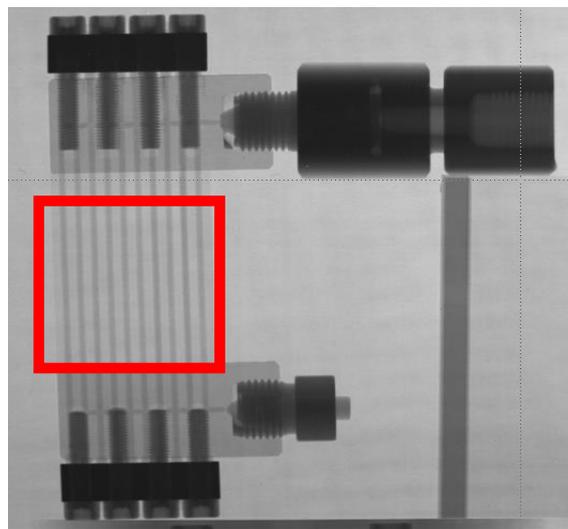
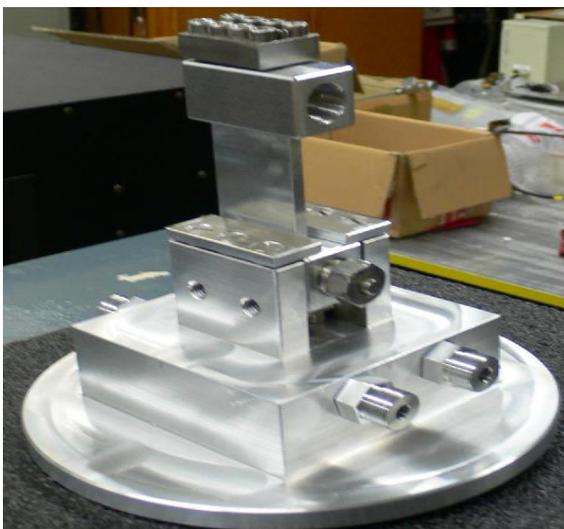
Design a new P cell for NSE

JCNS

Cell #2 --- improvement of performance

(Existing concept with Al7075 – 2kbar, R. Funer & J.Dörbecker)

3kbar, G. Simeoni)



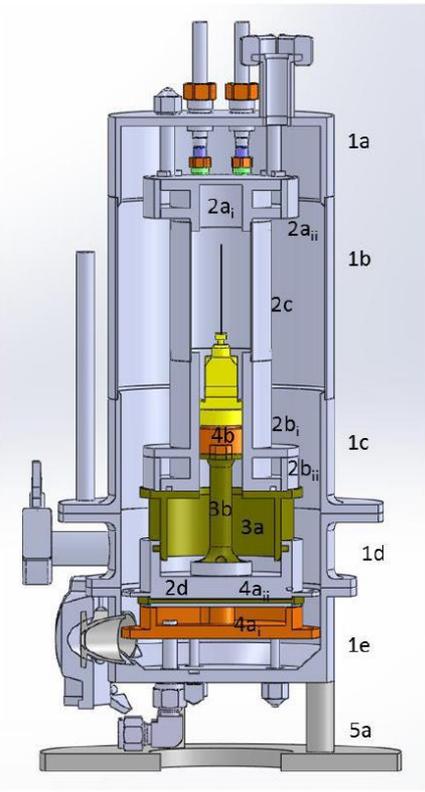
This concept with TiZr will lead to 5 (7) kbar !!! (H. Feilbach)

Concept already at ISIS for 7 kbar.

HZB, ILL

Design phase ✓

Final inner can



- Double walled (evacuated) Al
- Total size=400xØ110 (270 mm from base to sample)
- Inner cell=220xØ50 (170 mm above cold chamber region) , small volume for fast equilibration
- Wide angular scanning range possible (~300°), neutron windows with 15° opening
- 3 water chiller channels for precise temp.
- 2 resistive heating foils possible (below gonio and below reservoir)
- Sample thermally isolated (plastic post, steel supports)...

Manufacture ✓ 2 units (ILL, HZB)

Assembly ...

HZB



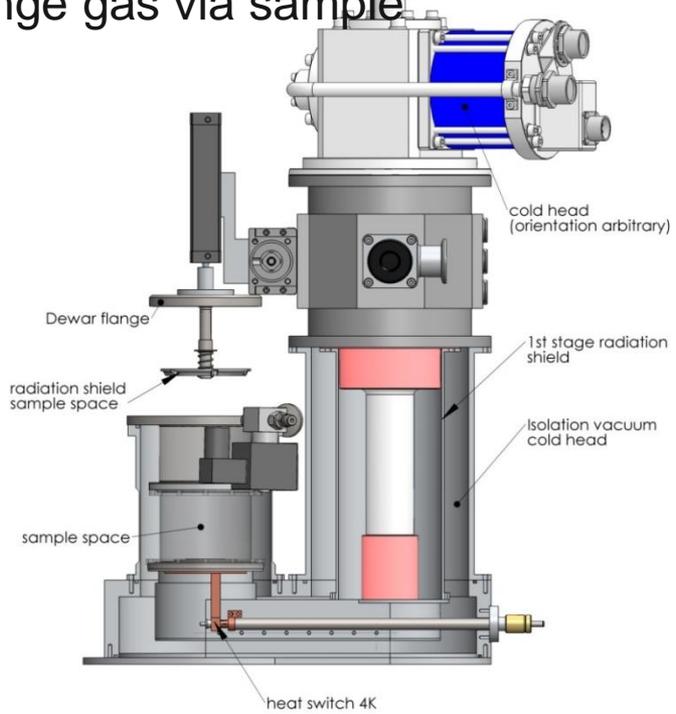
Delivery at ILL : week 38

1st experiments D16 (ILL) scheduled in November 2014

■ Compact cryostat

FRMII

- Separate sample space and cold head isolation vacuum
- Minimized cold mass
- Sample in exchange gas via sample container





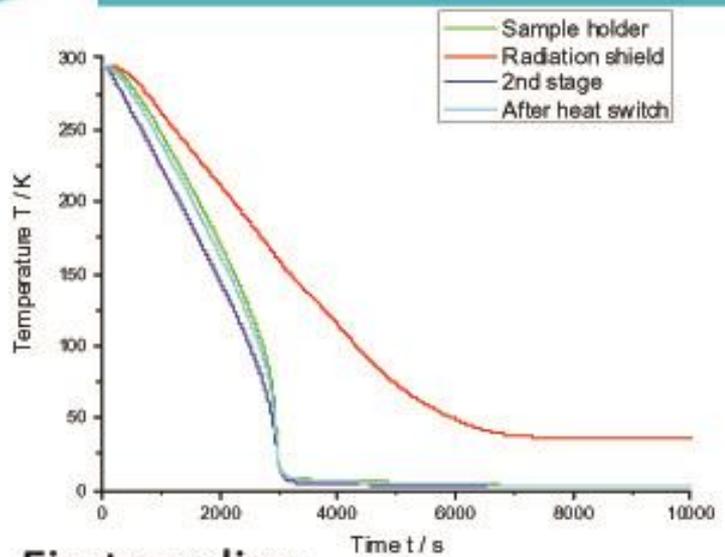
nmi3



Task 4

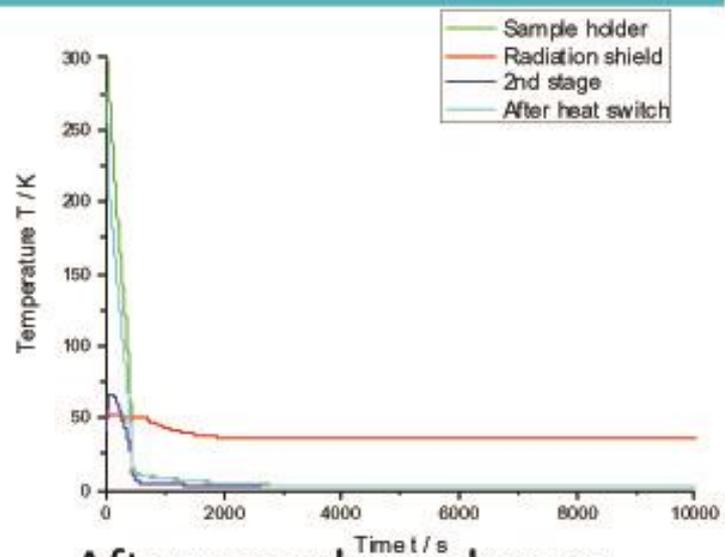
Cryogen-free cryostat with sample changer

Cooling performances 05 2014



First cooling

1:50 h: 2nd stage at ~2,8 K
 2:50 h: 2nd stage at ~2,7 K
 2:10 h: Sample at ~3,1 K
 $\Delta T \approx 0,4$ K



After sample exchange

0:40 h: 2nd stage at ~2,8 K
 0:55 h: 2nd stage at ~2,7 K
 0:50 h: Sample at ~3,4 K
 $\Delta T \approx 0,6$ K
 (0:10 h: sample at ~ 20 K)

- Robot for sample change under study ...

Next meeting:

- October 15-16 , 2014 Hosted by ISIS (STFC)
At the 8th International Workshop on Sample Environment
Oxfordshire, 12th– 16th October 2014
- May 2015 Saclay Hosted by LLB (CEA)

Change of MM distribution:

- From CEA ~2/3MM to ILL
to develop a prototype of SANS pressure cell
- Discussion on MM of JCNS about pressure cell sub-task.
Decision at the next JRA meeting, October 2014.