Laser-Polarized Xenon for NMR and MRI

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Laser-Polarized Xenon for NMR and MRI

- Production of hyperpolarized gas
- Polarization transfer from xenon to other nuclei ?
- ^{129}XeNMR-based biosensing
- Alternative detection method
- Perspectives

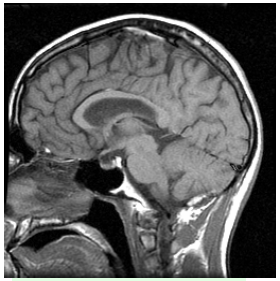
NMR

Method of low sensitivity (requires 10^{17}-10^{19} spins)

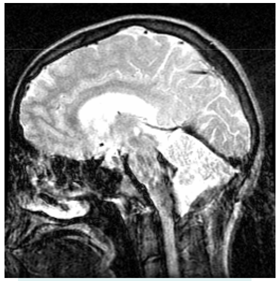
Ex: Imaging

- high spatial resolution
- good penetration depth
- multiparametric

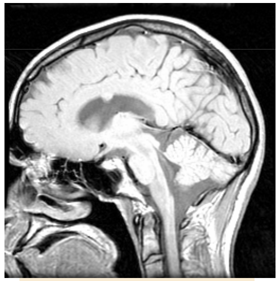
- low sensitivity



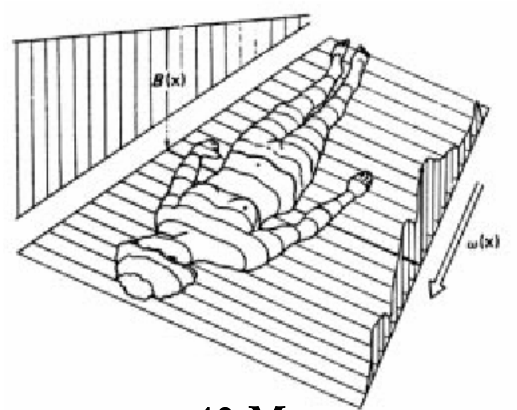
T_1 Contrast



T_2 Contrast

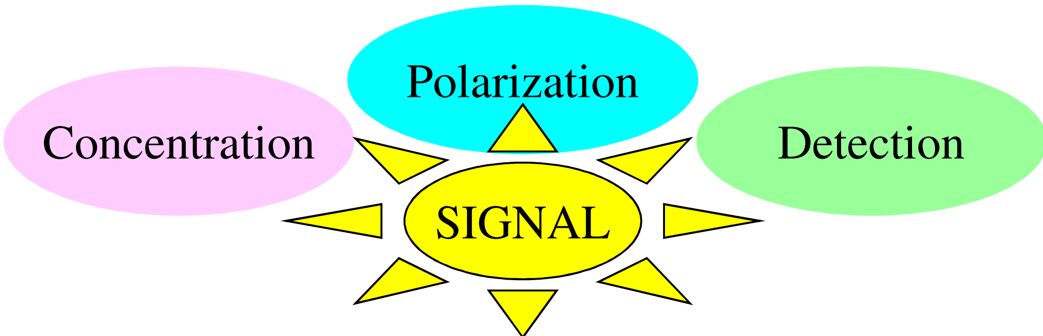


^1H Density



40 M

➔ 10^{17} spins per voxel of 100 x 100 x 400 μm^3



Nuclear Polarization

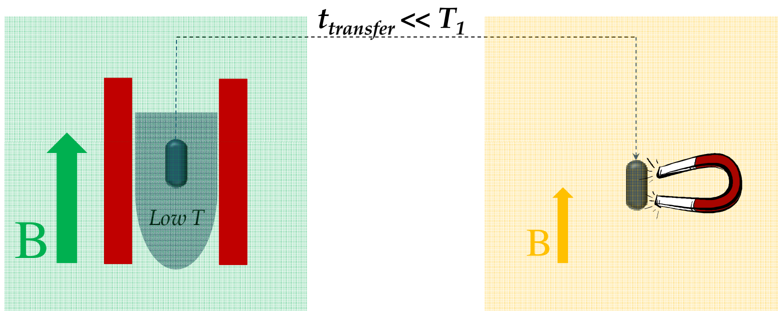
At Boltzmann equilibrium:

$$P = \frac{n_\alpha - n_\beta}{n_\alpha + n_\beta} = \tanh \underbrace{\frac{\gamma\hbar B_0}{2kT} \sim \frac{\gamma\hbar B_0}{2kT}}_{\textit{High T approx.}}$$

At 11.7 T (500 MHz ^{1}H) and 300K,

$P_{^1H} = 4\ 10^{-5}$

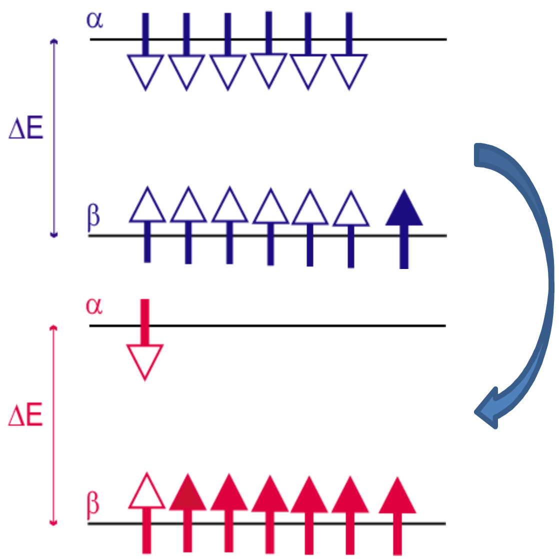
➔Brute force (pre-polarization method):



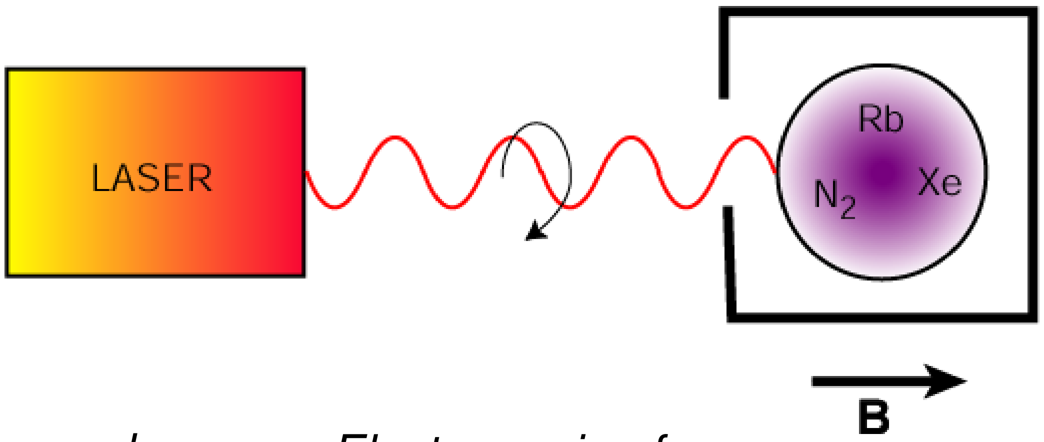
Pre-polarization *Study*

➔Polarisation transfer from a more ordered system:

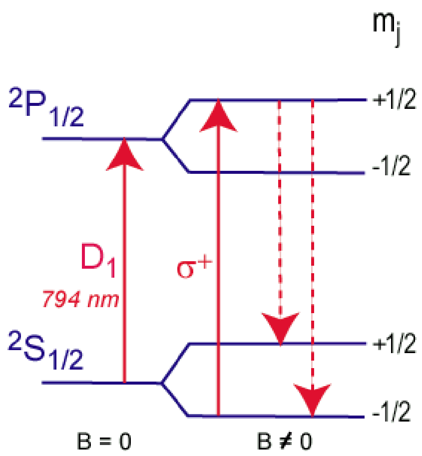
- electrons (DNP)
- parahydrogen (PHIP)
- photons (OP)

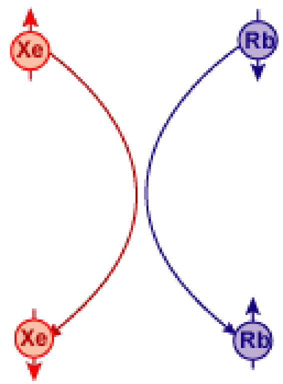


Noble gas optical pumping through spin-exchange method



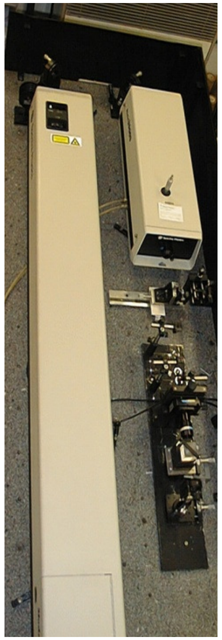
Photon angular momentum → *Electron spin of an alkali metal* → *Noble gas nuclear spin*

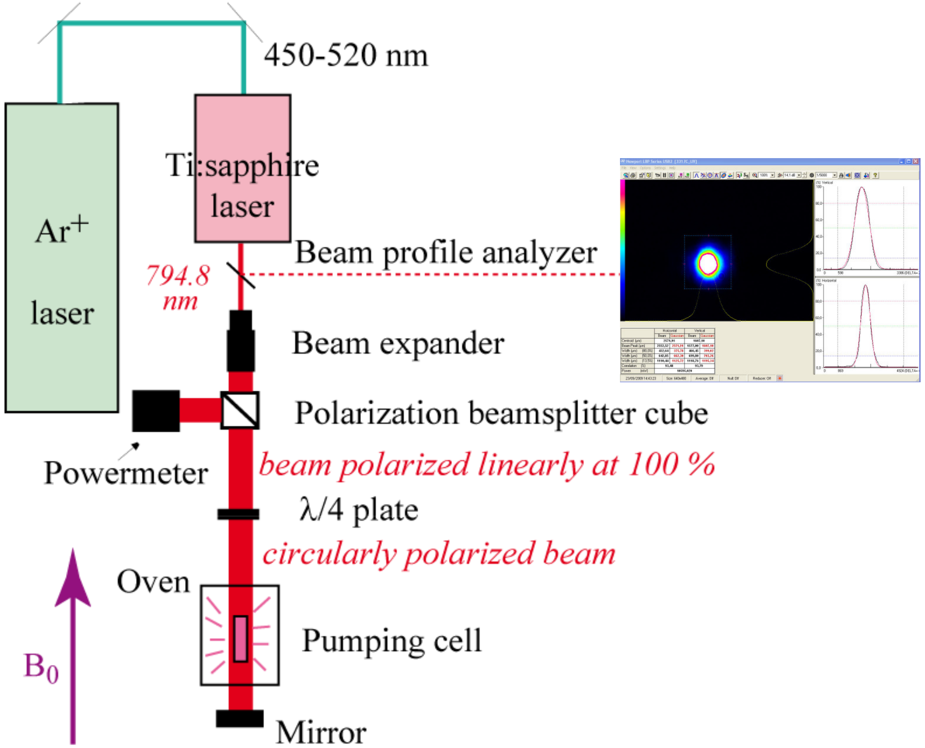


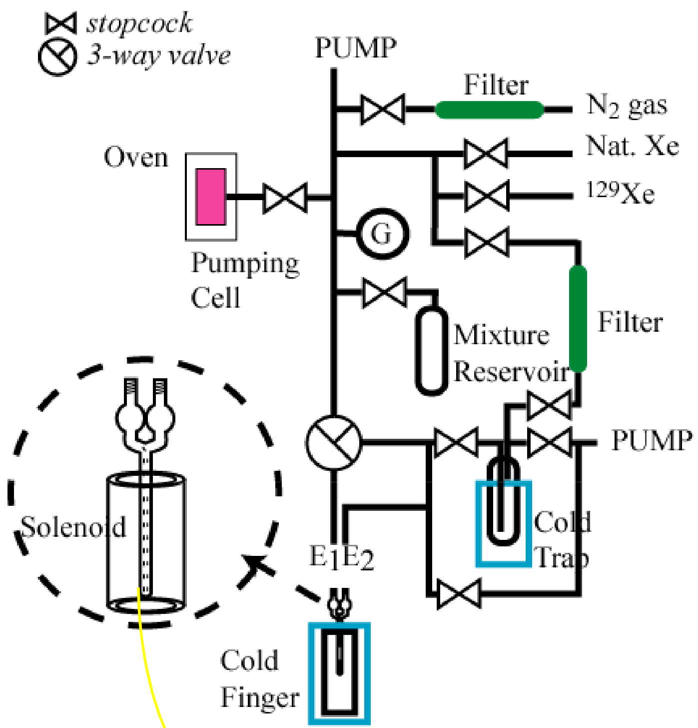


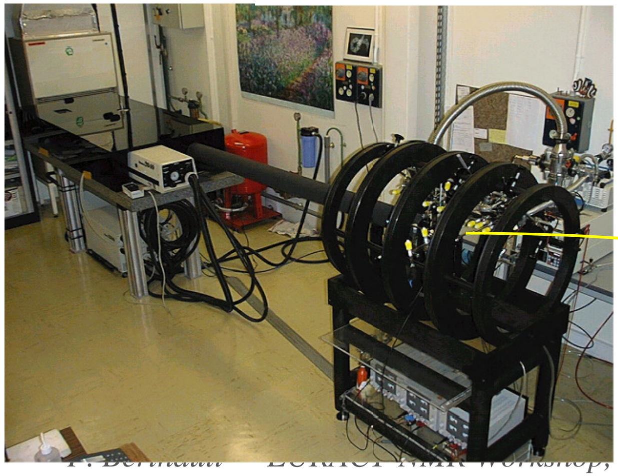
Walker and Happer, Rev. Mod. Phys., ***69*** *(1997) 629*

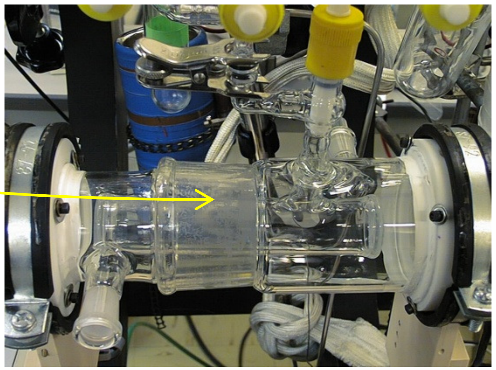
Experimental set-up

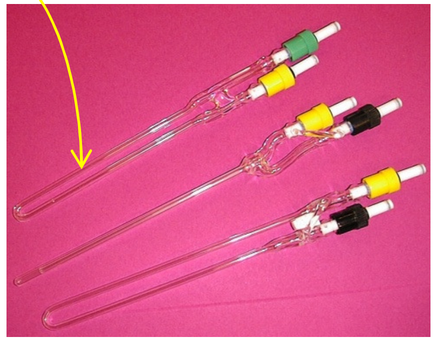








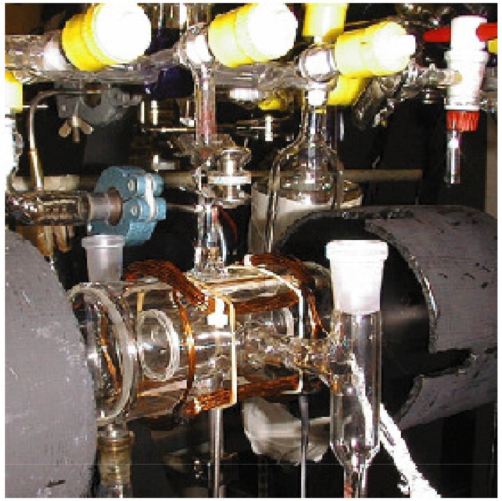


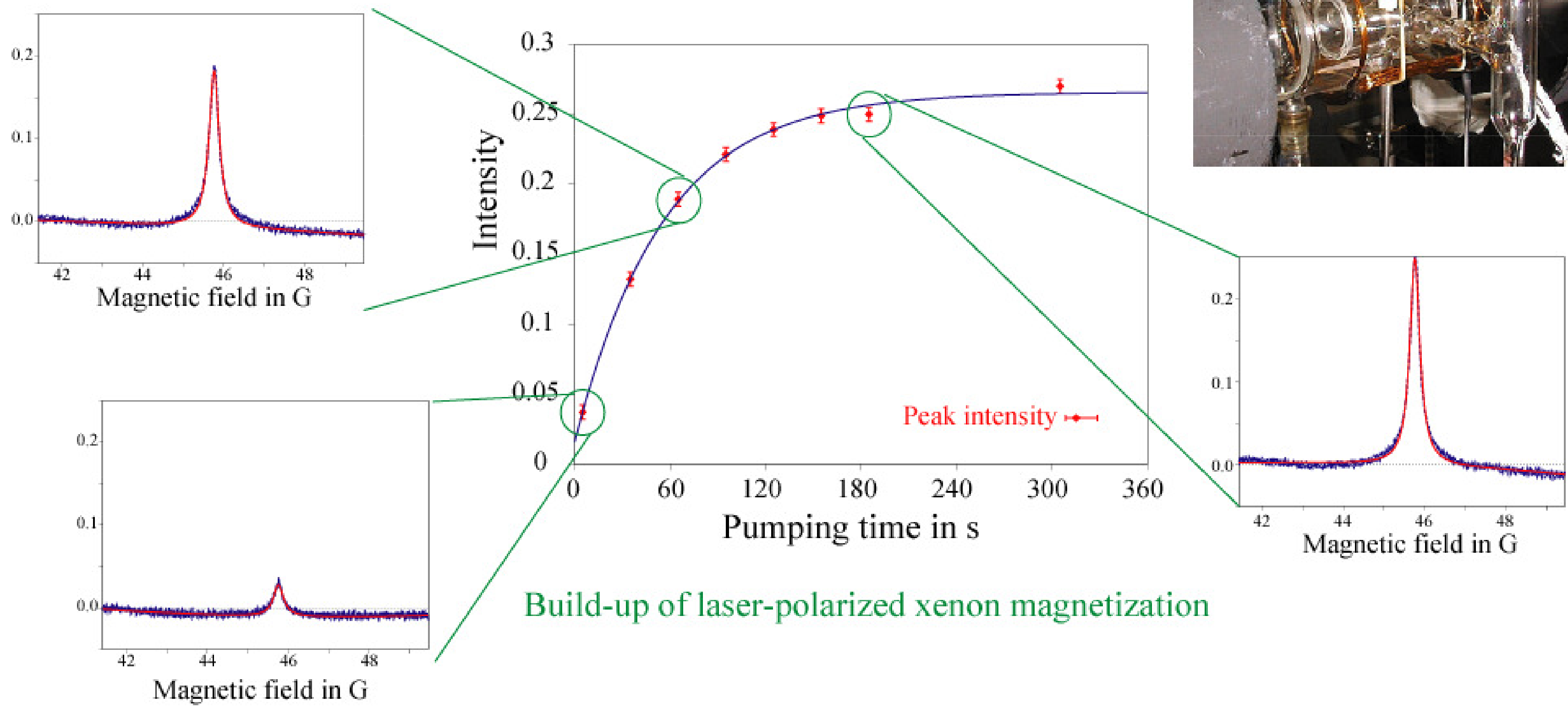


Eur. Phys. J. D, **12** (2000) 289

Monitoring of the Xe polarization via an *in situ* CW spectrometer

* Working frequency : 53.1 kHz
* Adiabatic fast passage
* Detection by crossed coils
* Controlled by a PC

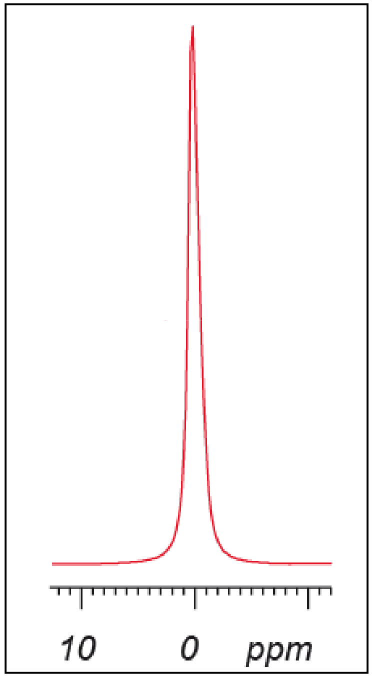




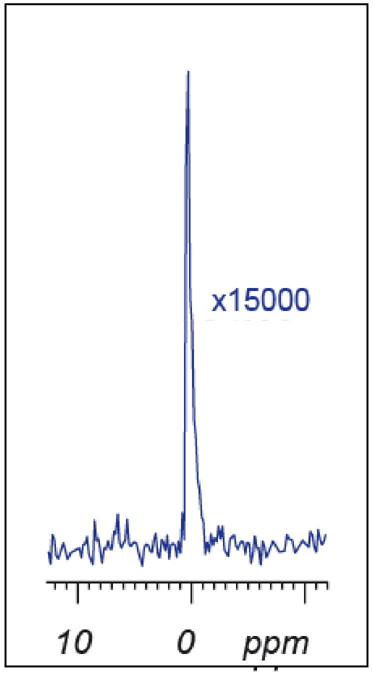
Build-up of laser-polarized xenon magnetization

Laser-Polarized Xenon

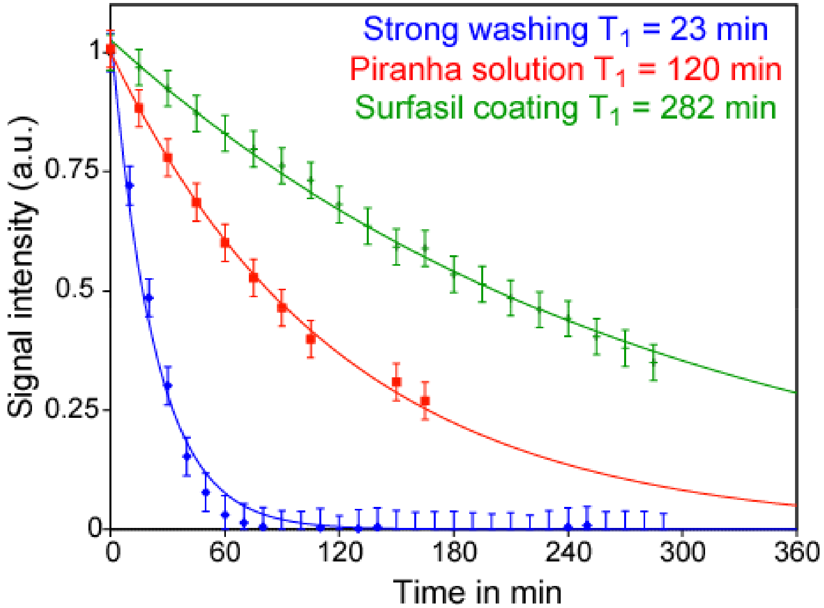
Typical xenon signal enhancement: > 40 000



Laser polarized xenon
1.1 Atm
11.7T, 290K, 1 scan
Polarization : **50%**



Thermal equilibrium signal
Same sample
11.7T, 290 K, 32 scans
15h of acquisition



Relaxation at 290 K in a magnetic field homogeneous in direction (5-11.7 T)

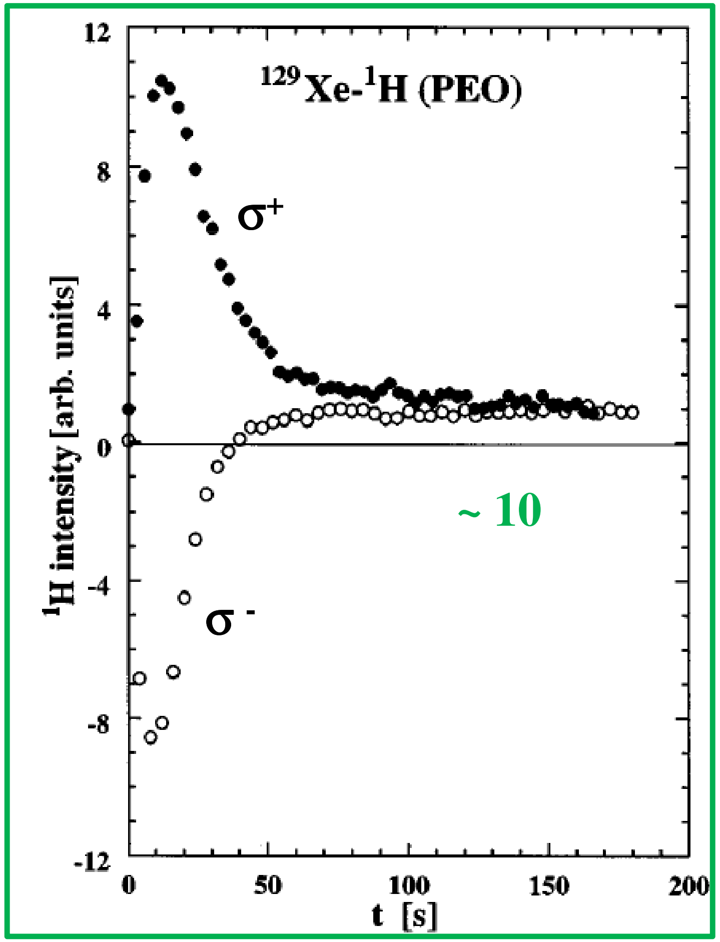
Rate of production : 1 mL under 1 atm in 5 min

Magnetization transfer from polarized xenon

^{129}Xe has few ways to give usefully its magnetization (saturated electron layers, spherical symmetry, large vdW radius, …).

Transfer methods:

- Nuclear Overhauser Effect (SPINOE)
  *Navon et al., Science **271** (1996) 1848*
- Hartmann-Hahn cross-polarization
  *Long et al., J. Am. Chem. Soc. **115** (1993) 8491*
- Thermal mixing at low field
  *Bowers et al., Chem. Phys. Lett. **205** (1993) 168*



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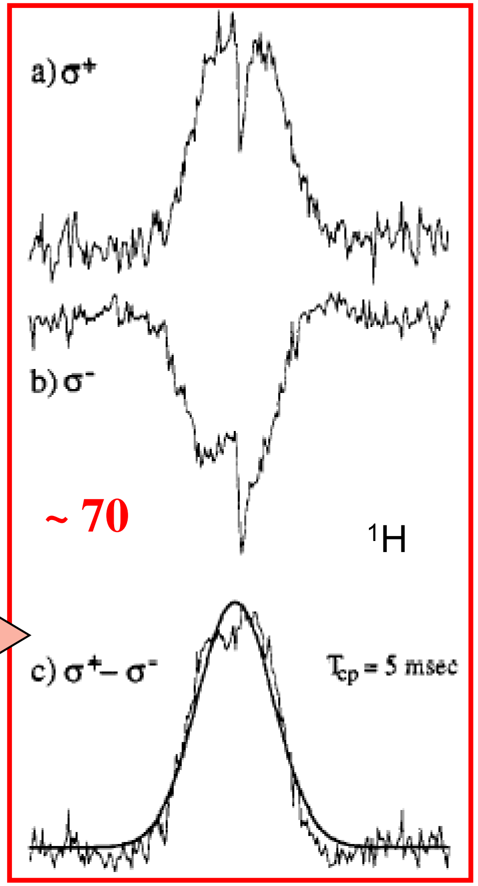

Microporous polymer
Poly(triarylcarbinol)

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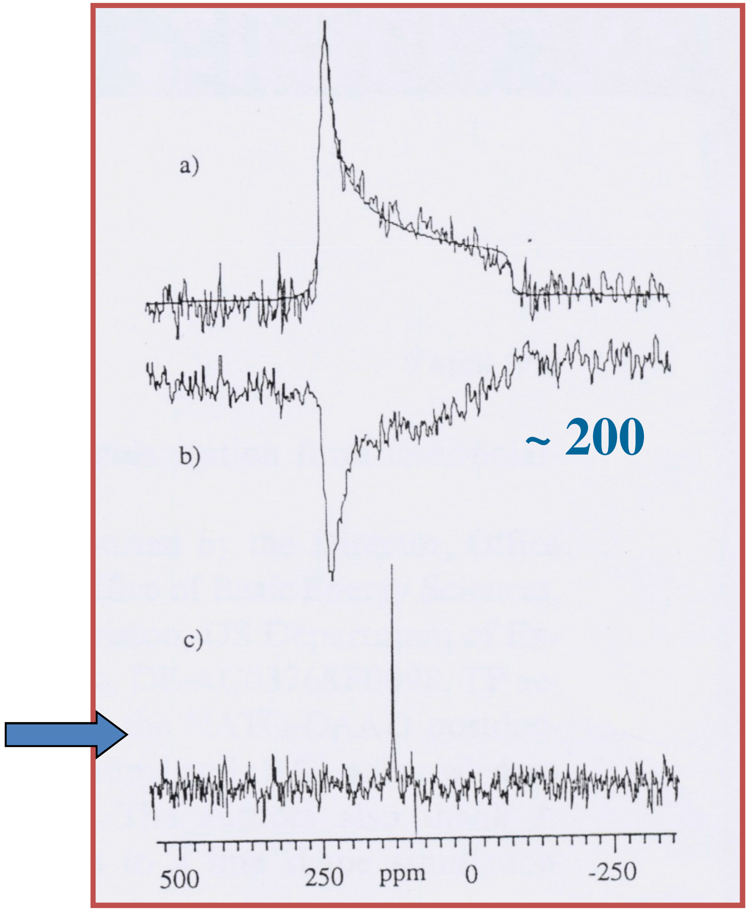


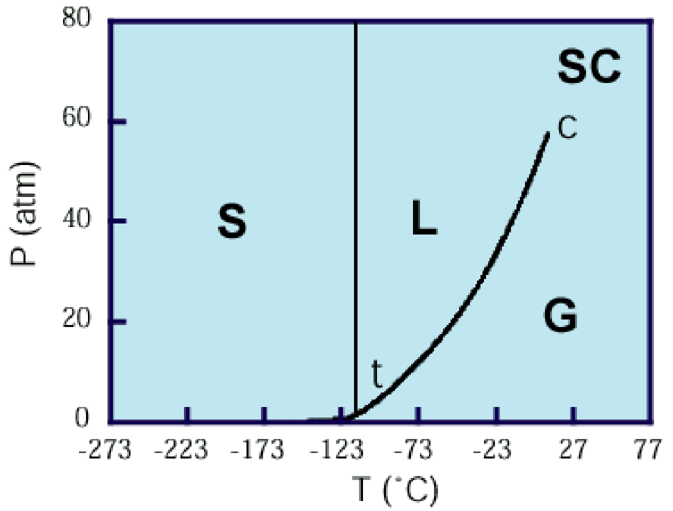
Xenon for NMR

• Starting idea: find an inert molecule, sensitive to physical interactions with other species and detectable through NMR, as a probe of molecular environment.

• Ideal probe !

- *monoatomic noble gas*
- *big electron cloud → each distortion of this cloud has immediate effects on NMR parameters*
- *2 isotopes easily observable :*
  - * ^{129}Xe *(spin 1/2) nat. ab. 26.4%*
  - * ^{131}Xe *(spin 3/2) nat. ab. 21.2%*
- *pronounced hydrophobic character*

Possibility to considerably increase its nuclear polarization, to use it easily under gaseous, dissolved, liquid, and even supercritical forms!
$P_c = 58$ atm ; $T_c = 17°C$



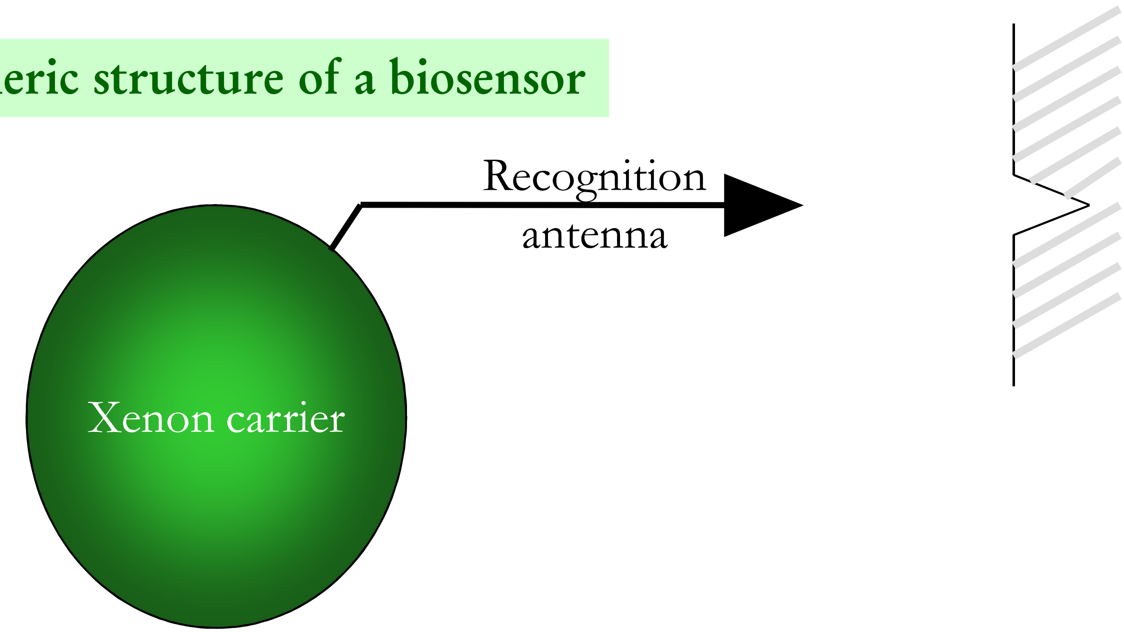
Biosensing Using Laser-Polarized Xenon NMR

Final goal: propose a sensitive MR imaging for early detection of biological events

Tool: NMR of ^{129}Xe, whose nuclear polarization has been enhanced by 4-5 orders of magnitude via optical pumping

Requisite: xenon must be transported to the desired biological receptors before imaging

Generic structure of a biosensor



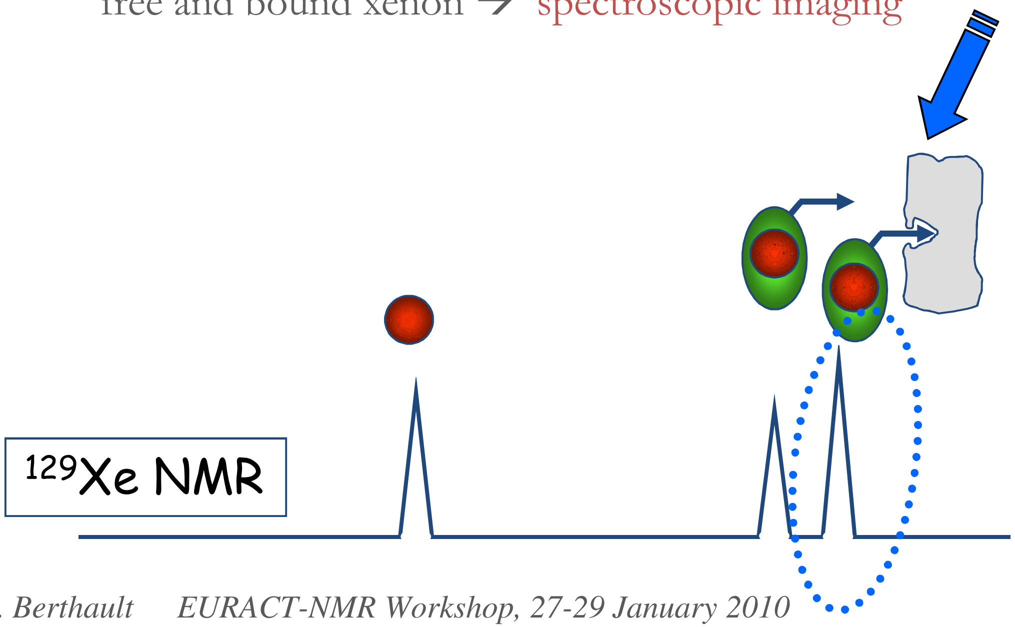
Biosensing Using Laser-Polarized Xenon NMR

- **First approach**

  The host system transports many xenon atoms (tracer) and a density image is performed : poor efficiency (contrast)

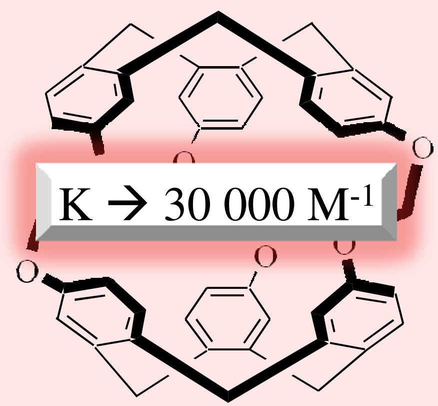
- **Second approach** *(Pines et al.)*

  Large polarizability of the xenon electron cloud → strong chemical shift effect → distinction on the ^{129}Xe spectrum between free and bound xenon → 'spectroscopic imaging'

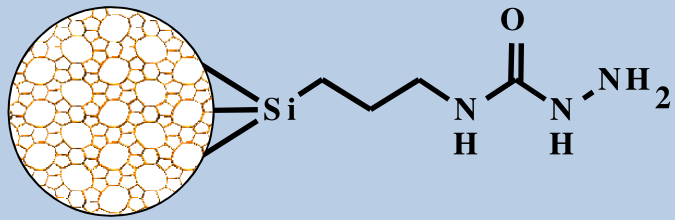


Choice of the xenon host

- High affinity for the noble gas / high number of atoms transported
- Slow relaxation of encapsulated xenon
- Functionalizable by biological ligands
- Various xenon chemical shifts in different host systems
- Xenon chemical shift variation when the biosensor is bound to its target
- Xenon in-out exchange



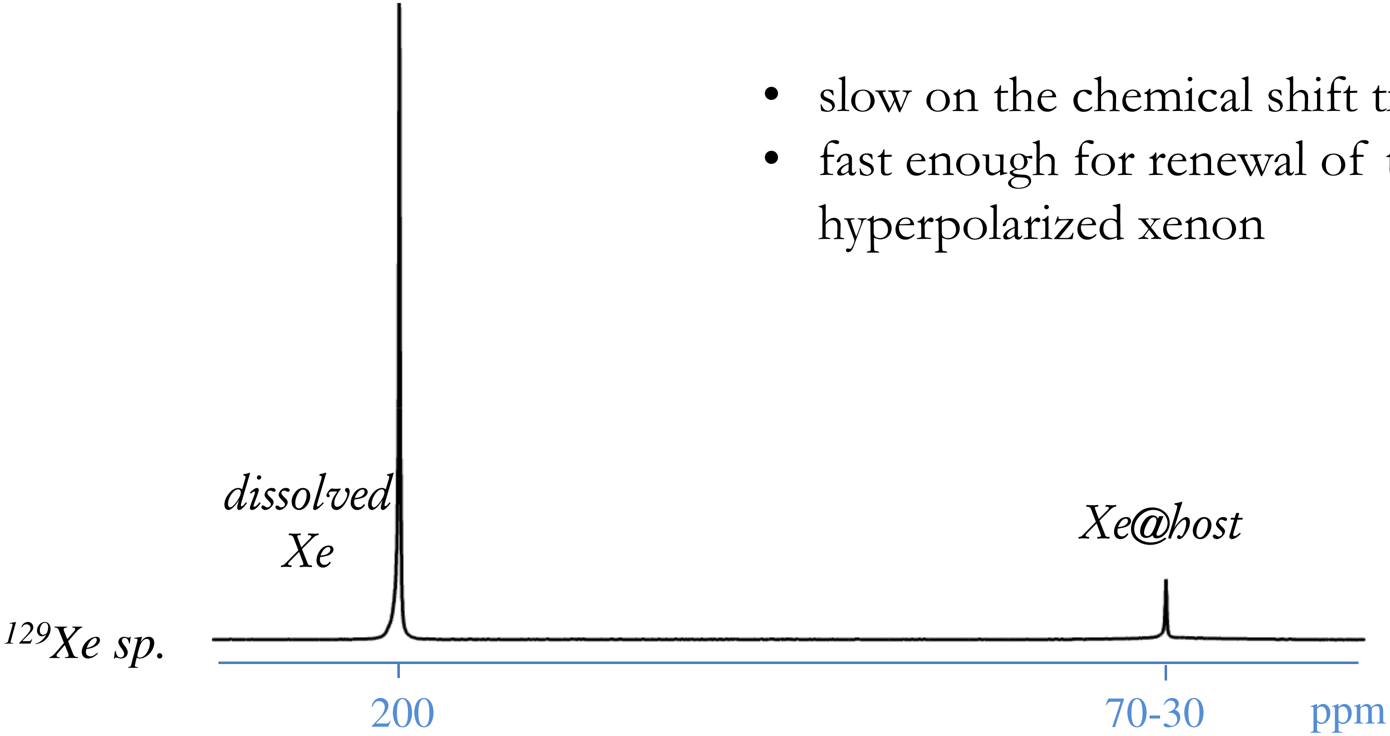
Cryptophanes

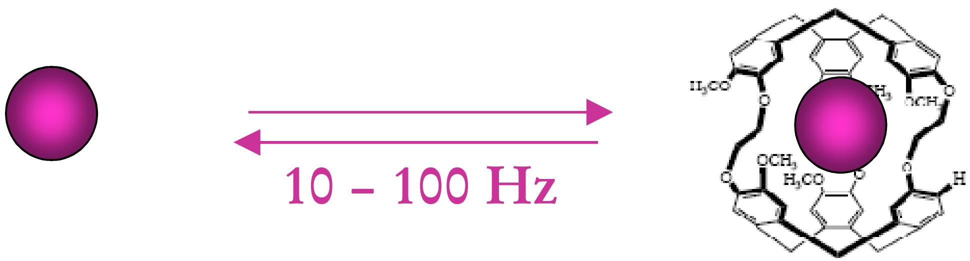


Functionalized Microporous Colloids

Xenon in-out exchange

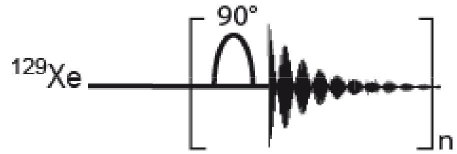
- slow on the chemical shift time scale
- fast enough for renewal of the cage with hyperpolarized xenon





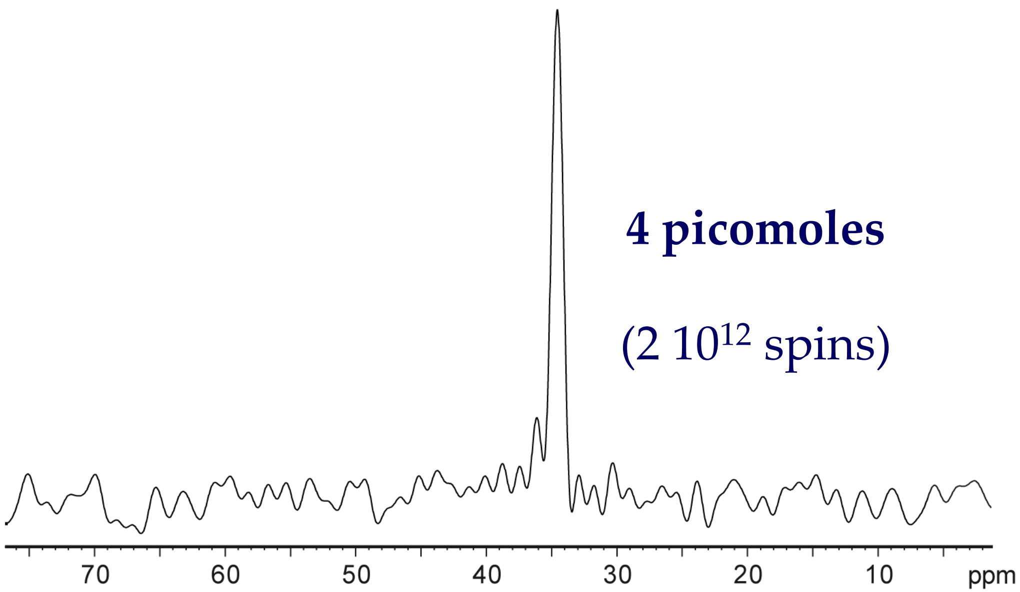
Xenon in-out exchange

➔Important gain in sensitivity



Solution of **12.8 nM** of cryptophane at 11.7 T and 310K.

n=3600 ; experiment time = 154 s.



4 picomoles

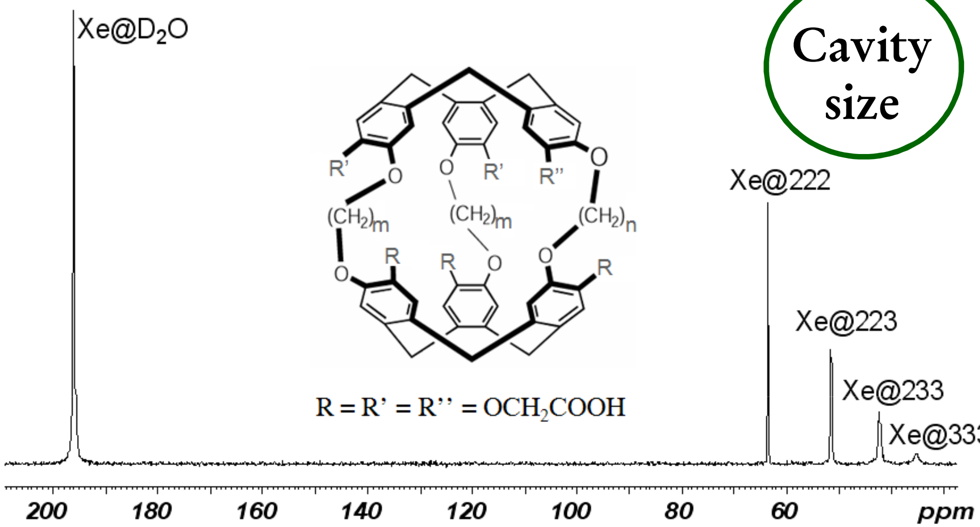
(2 10^{12} spins)

Prog. NMR. Spectrosc., ***55*** *(2009) 35*

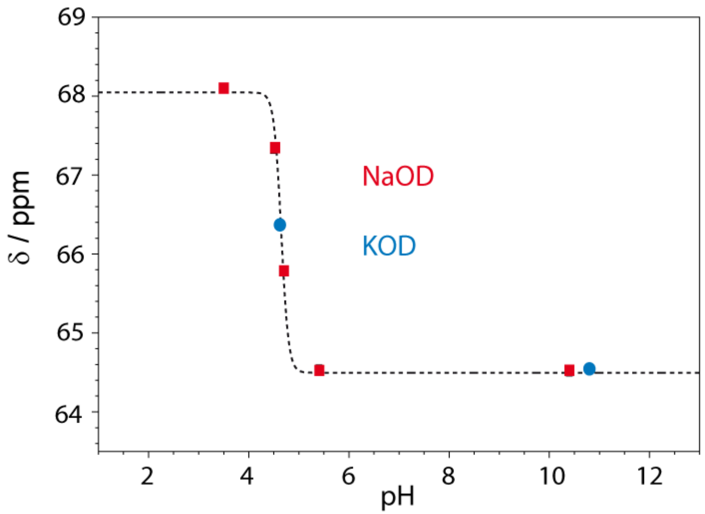
Multiplexing capabilities

$\delta_{Xe@host}$ **may depend on:**

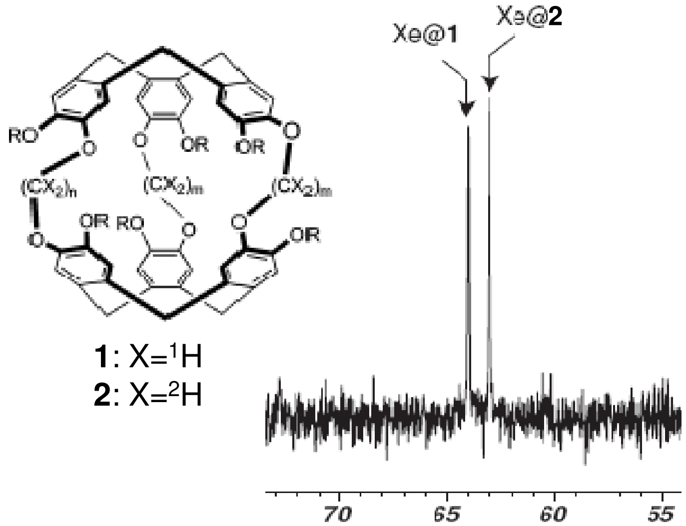
Cavity size



pH



Host substituents

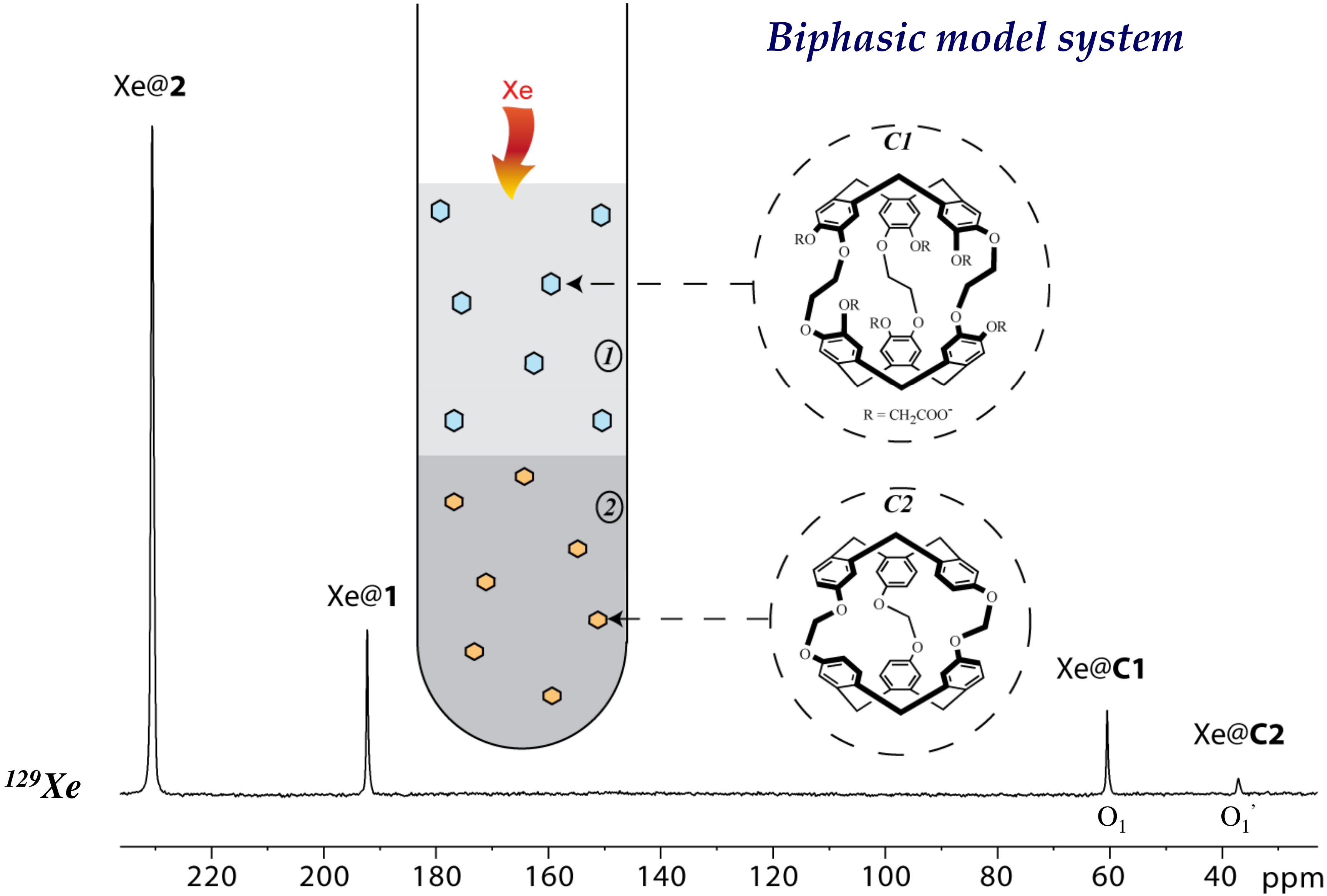


Interest: simultaneous detection of correlated biological events

J. Am. Chem. Soc. ***128*** *(2006) 6239*

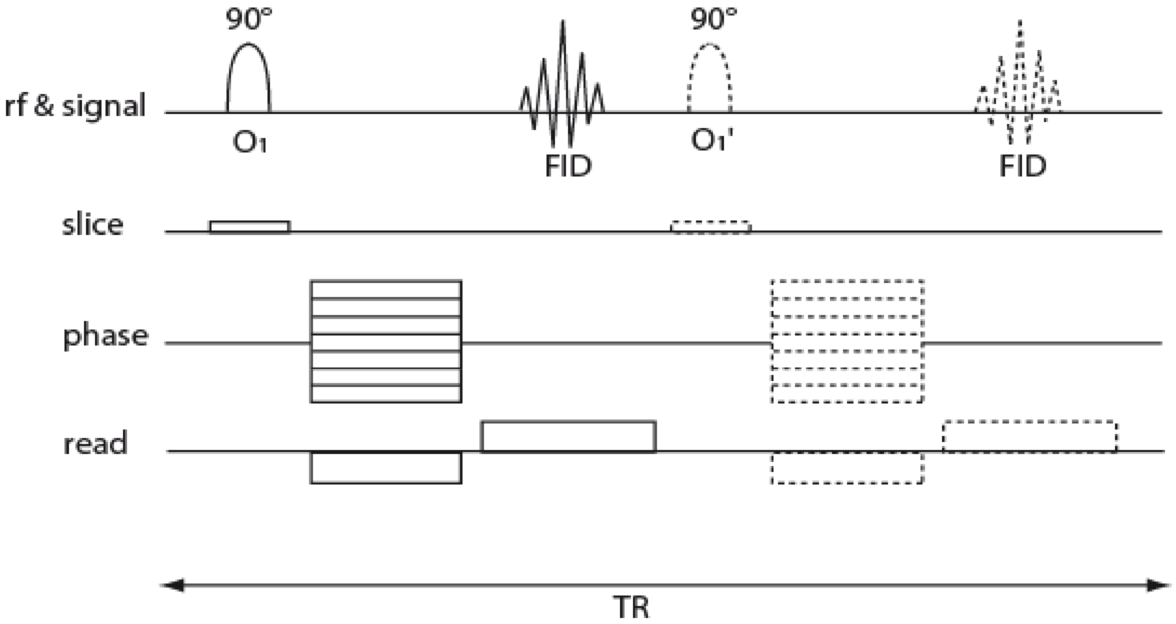
Fast multiplexed imaging

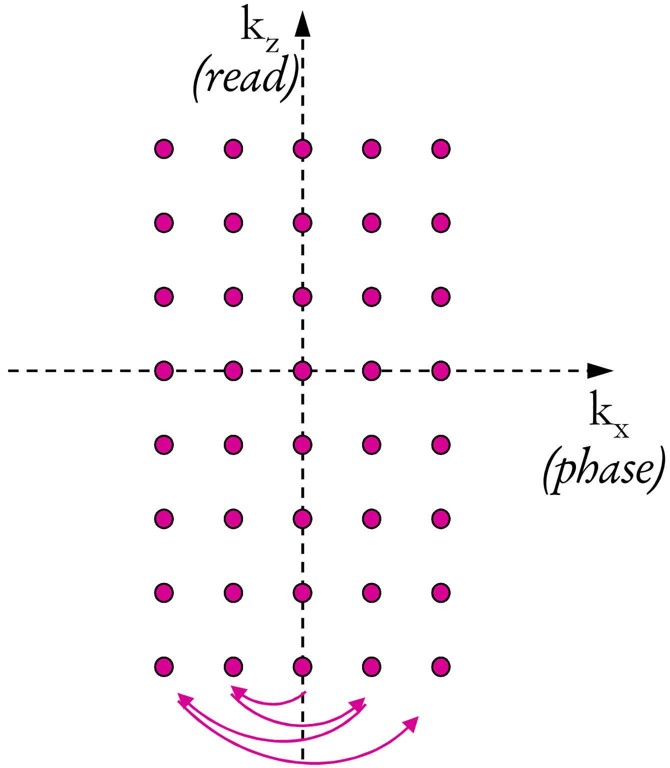
Biphasic model system



Fast multiplexed imaging

Used MRI sequence





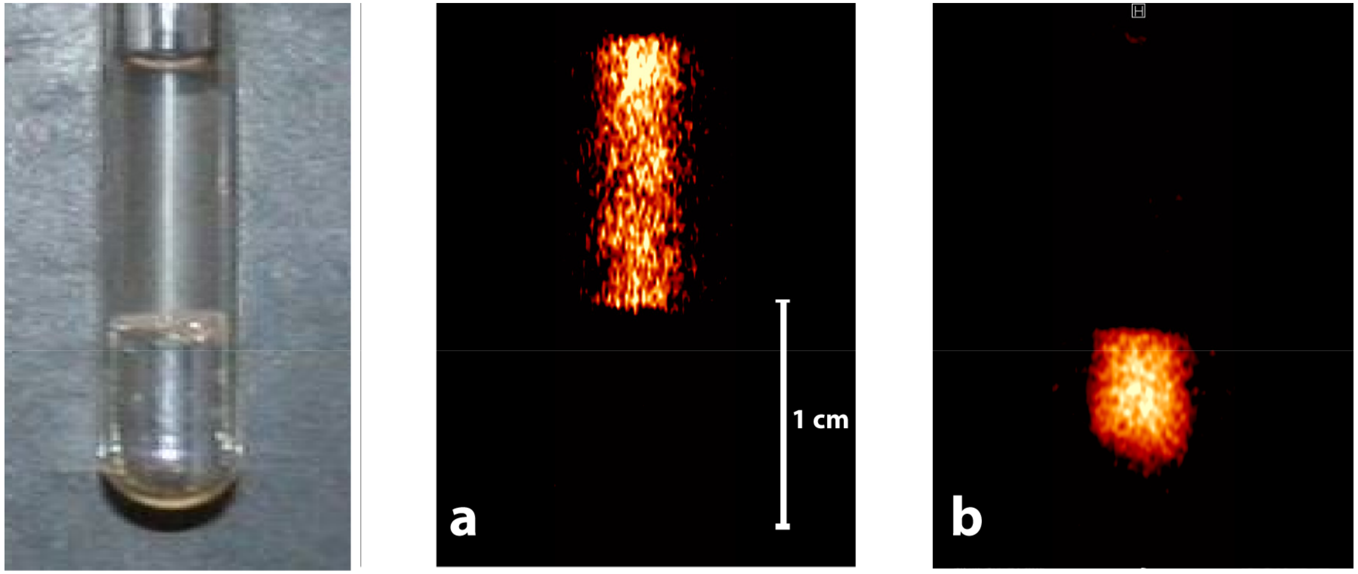
- Cartesian sampling of the reciprocal *k* space
(frequency selective gradient echo)
- Interleaved acquisition with offset switch
- Centrically ordered gradients for the phase dimension

$\{0\ ;\ -G_{min}\ ;\ +G_{min}\ ;\ \ldots\ ;\ -G_{max}\ ;\ +G_{max}\}$

Alternative:
HYPERCEST

Fast multiplexed imaging

Results



Concentrations and volumes:
$[C1]=300\ \mu M$; $V_{C1} = 300\mu L$
$[C2]=300\ \mu M$; $V_{C2} = 200\mu L$

Experiment time : **25 s** (TR = 50 ms)
Resolution : 117 µm x 125 µm

10^{13} spins per voxel

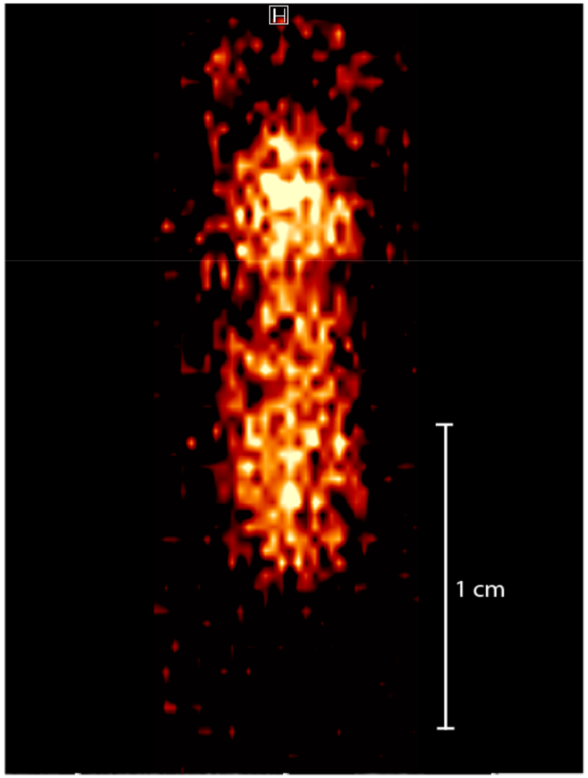
J. Am. Chem. Soc., ***130*** *(2008) 16456*

Pushing the sensitivity limits

[C2] = 25 μM
V_{C2} = 500μL

Experiment time : 204 s
Resolution : 117 μm x 250 μm

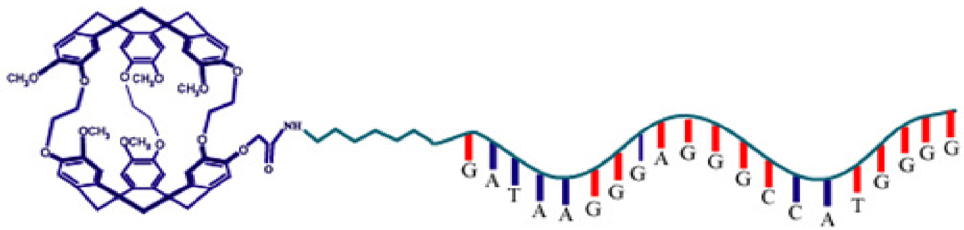
2.10^{12} spins per voxel



J. Am. Chem. Soc., ***130*** *(2008) 16456*

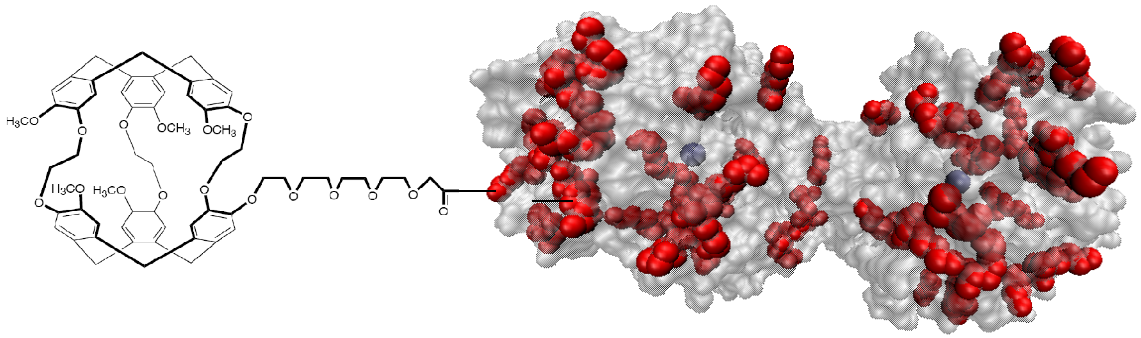
Our First Biosensors

1



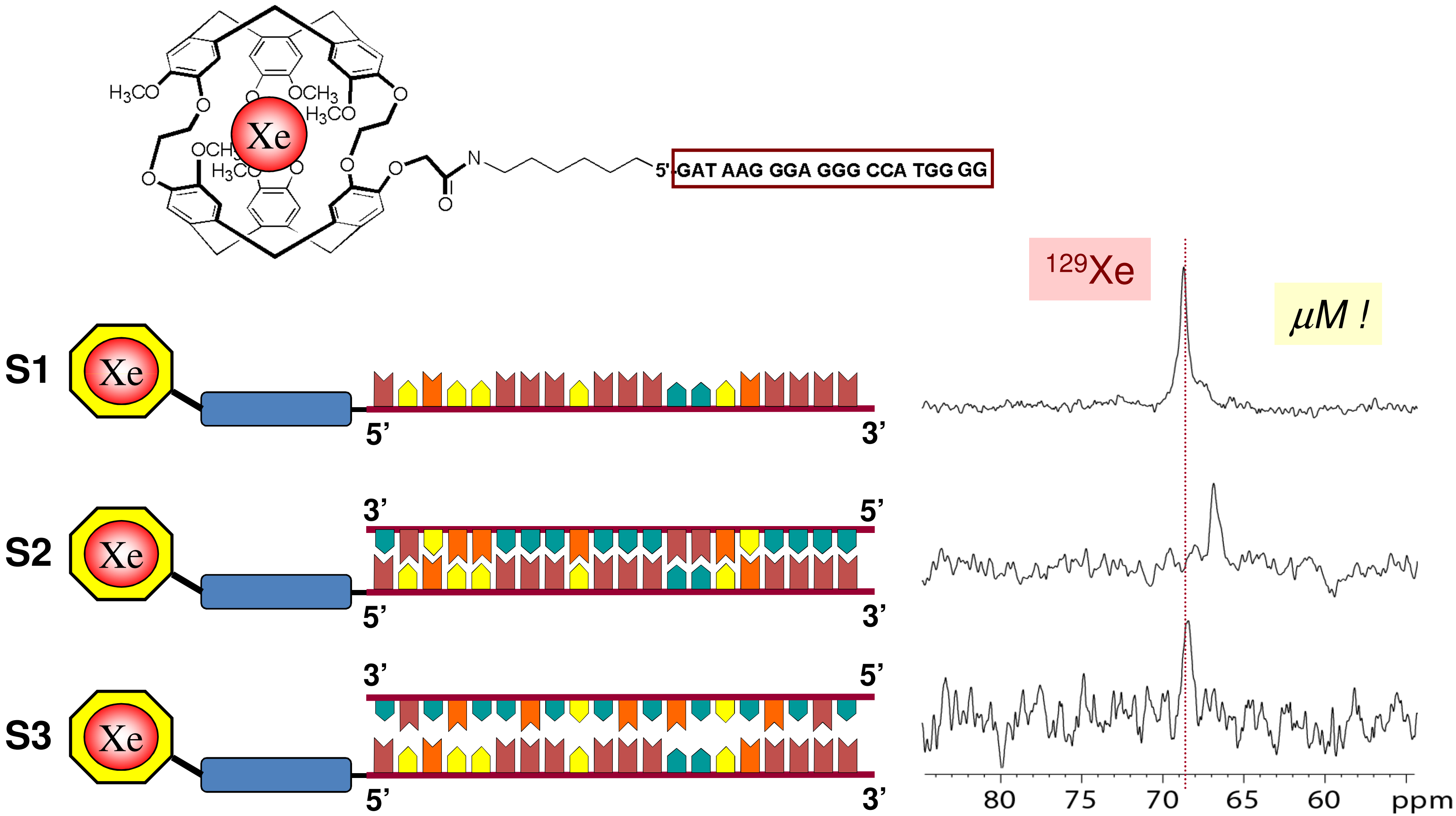
Recognition of DNA strands

2



Transferrin labelling

In vitro recognition of a DNA strand

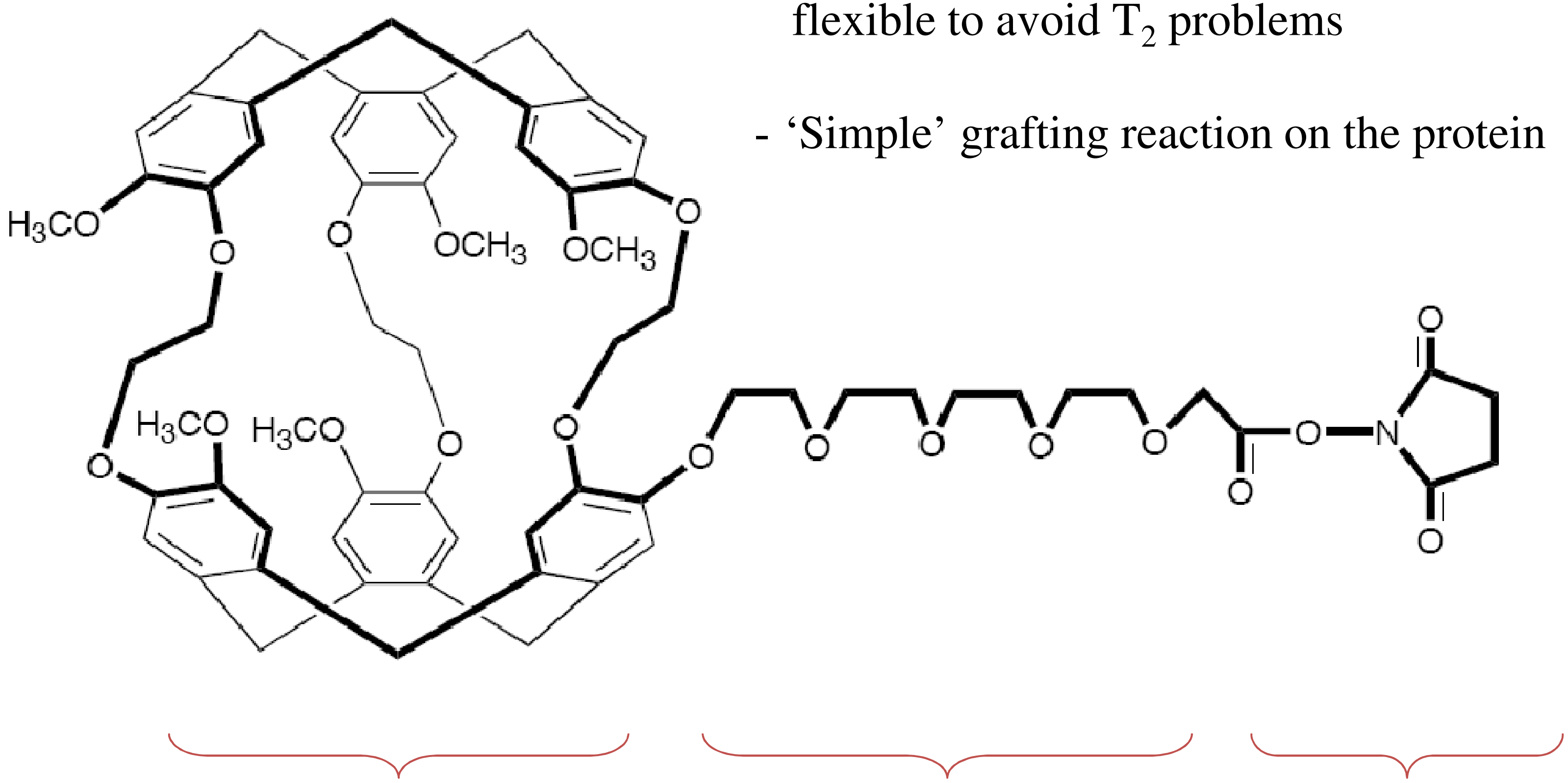


ChemPhysChem **8** *(2007) 2082*

Transferrin system : Precursor for a biosensor

- Spacer long enough to avoid the multiplication of ^{129}Xe peaks due to racemization, and flexible to avoid T_2 problems

- ‘Simple’ grafting reaction on the protein



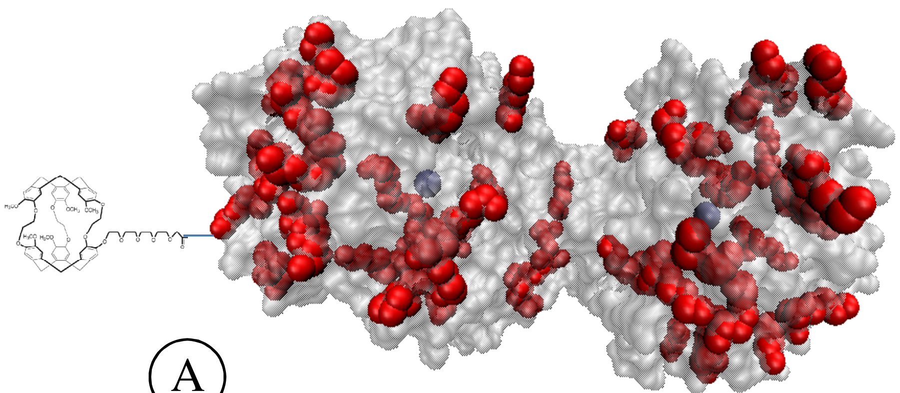
Cryptophane *PEG* *Succinidimidic group*

Choice of the transferrin system for *in vitro* assays

- Cell surface receptors
- Availability of cells with high number of receptors (K562)
- Endocytosis

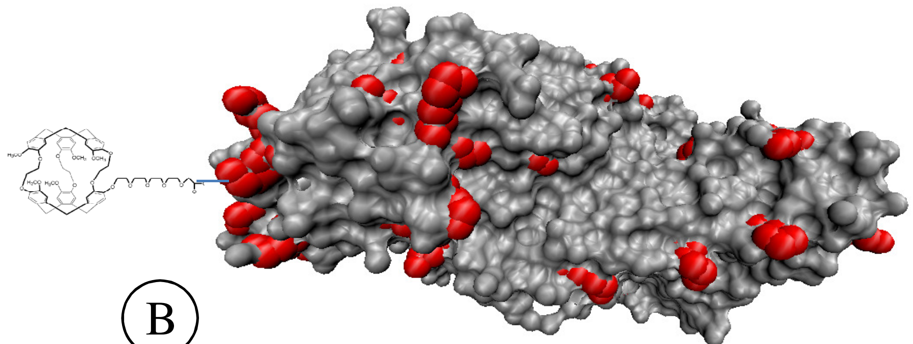
$\left.\right\}$ 10^5 targets per cell

- Affinity of holo-Tf = 700 x apo-Tf (and Fe can be replaced by Ga)
- Labelling of a big protein (80 kDa) by a small molecule
- Non specific grafting on the Lys residues → till 5 cryptophanes / Tf
- Possibility to compare with other biosensors easily built



A

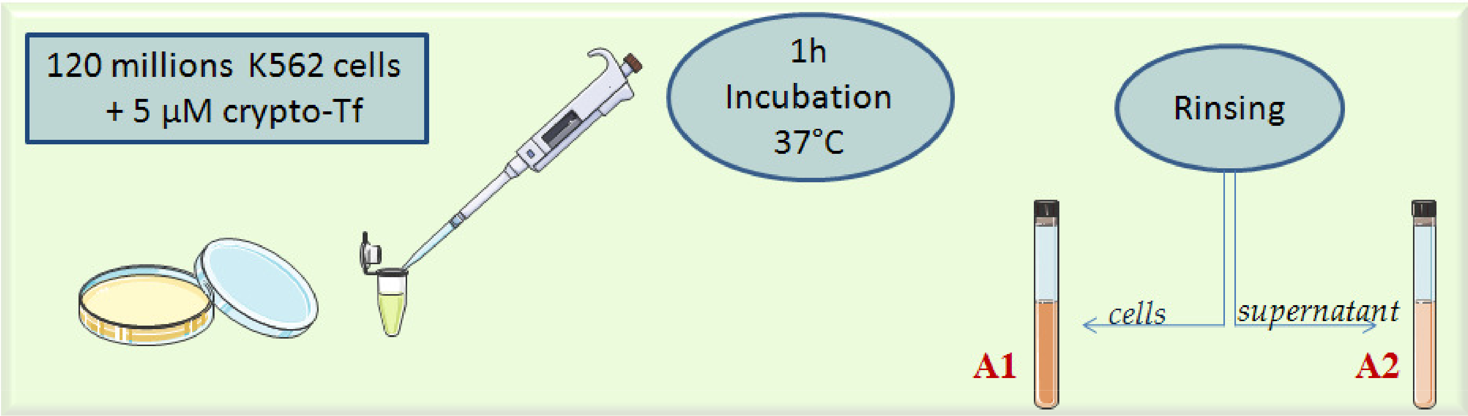
Cryptophane - Tf

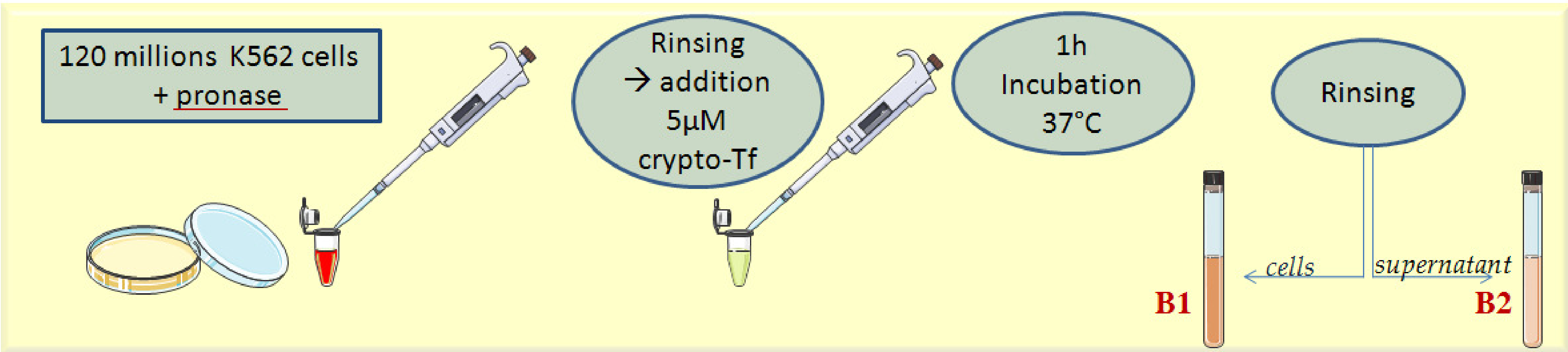


B

Cryptophane - BSA

Transferrin system:
Sequence of the first experiments

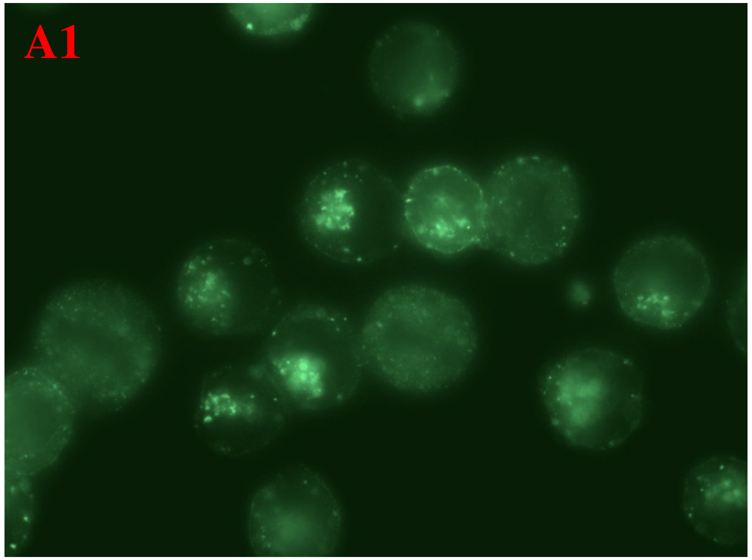


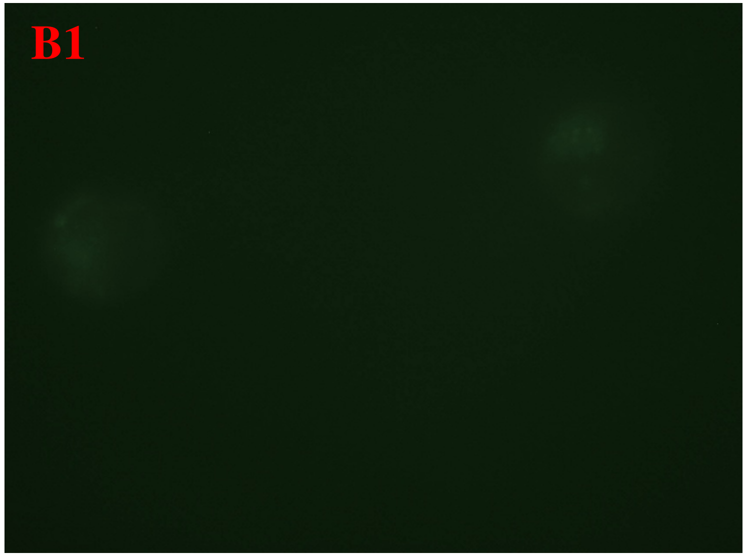


Transferrin system:
Fluorescence results

With pronase (proteolytic enzyme inactivating membrane receptors) at 2mg/mL

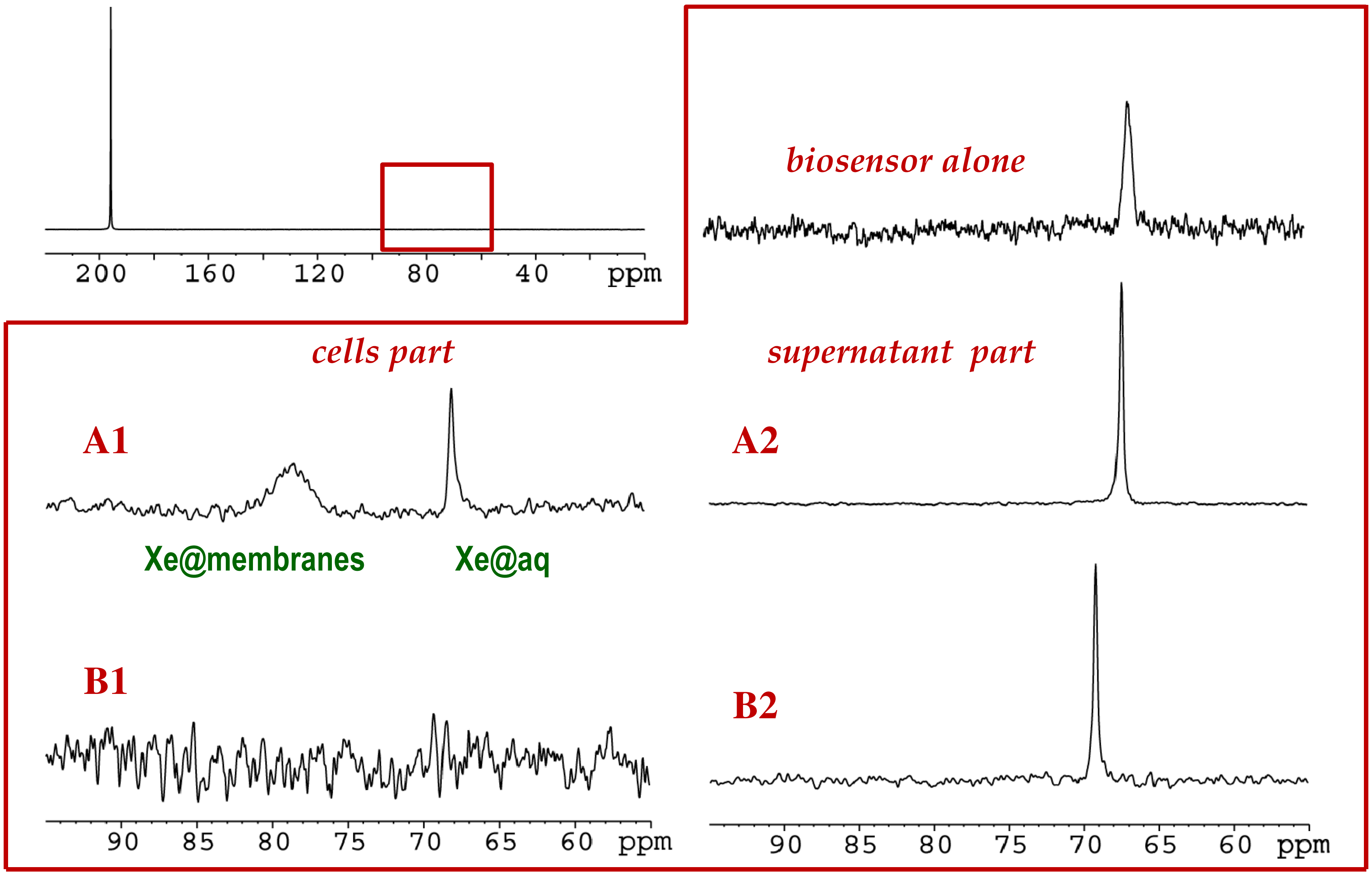
Cryptophane-Rhodamine G-Transferrin
R=2, R=2
37°C



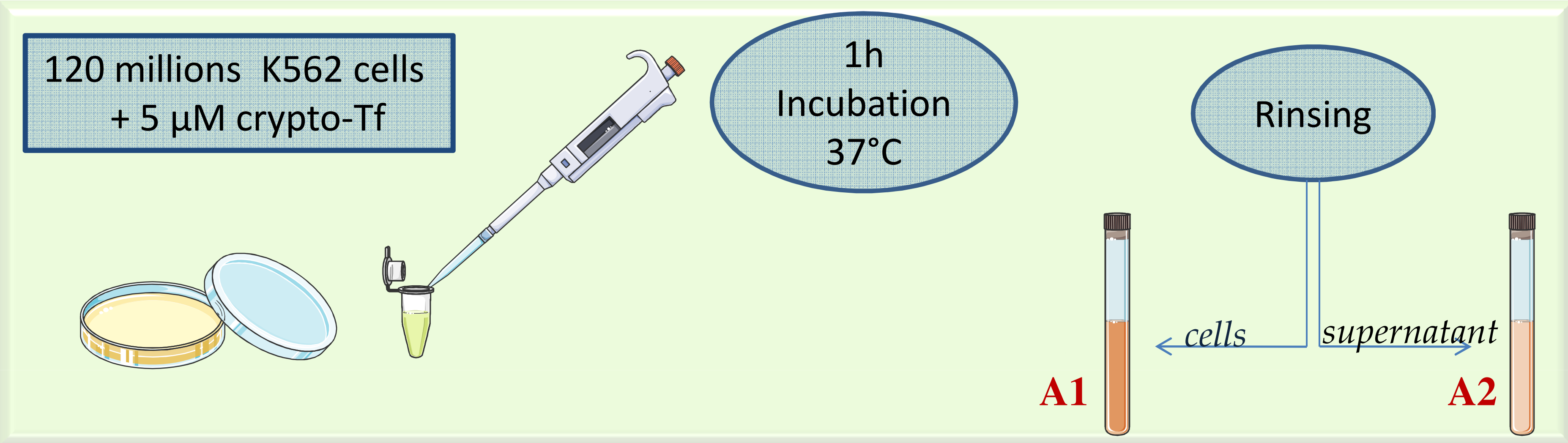


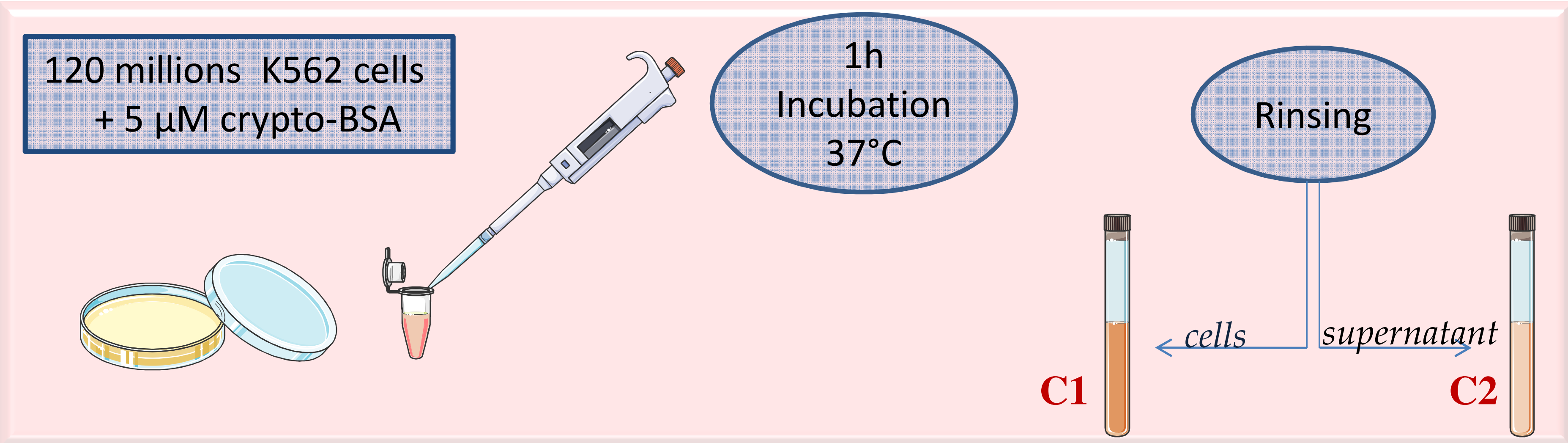
Incubation 1h, concentration 200nM

Transferrin system:
^{129}Xe NMR results



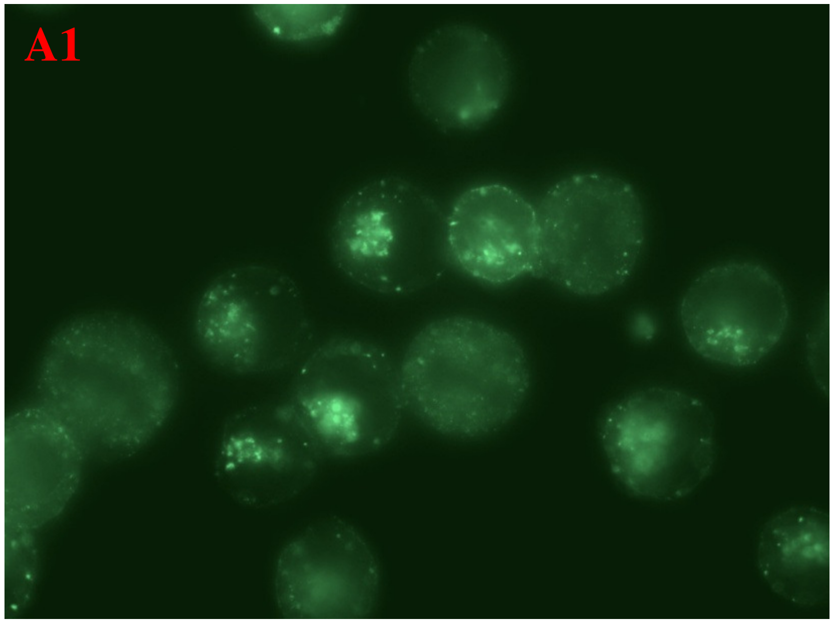
Transferrin system:
Sequence of the second experiments



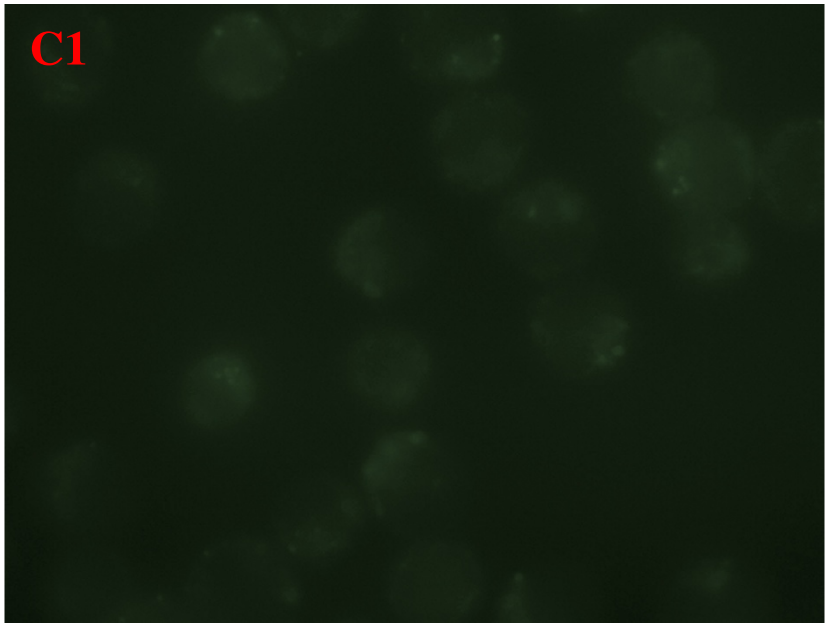


Transferrin system:
Fluorescence results

Cryptophane-
Rhodamine G-
Transferrin
R=2, R=2
37°C

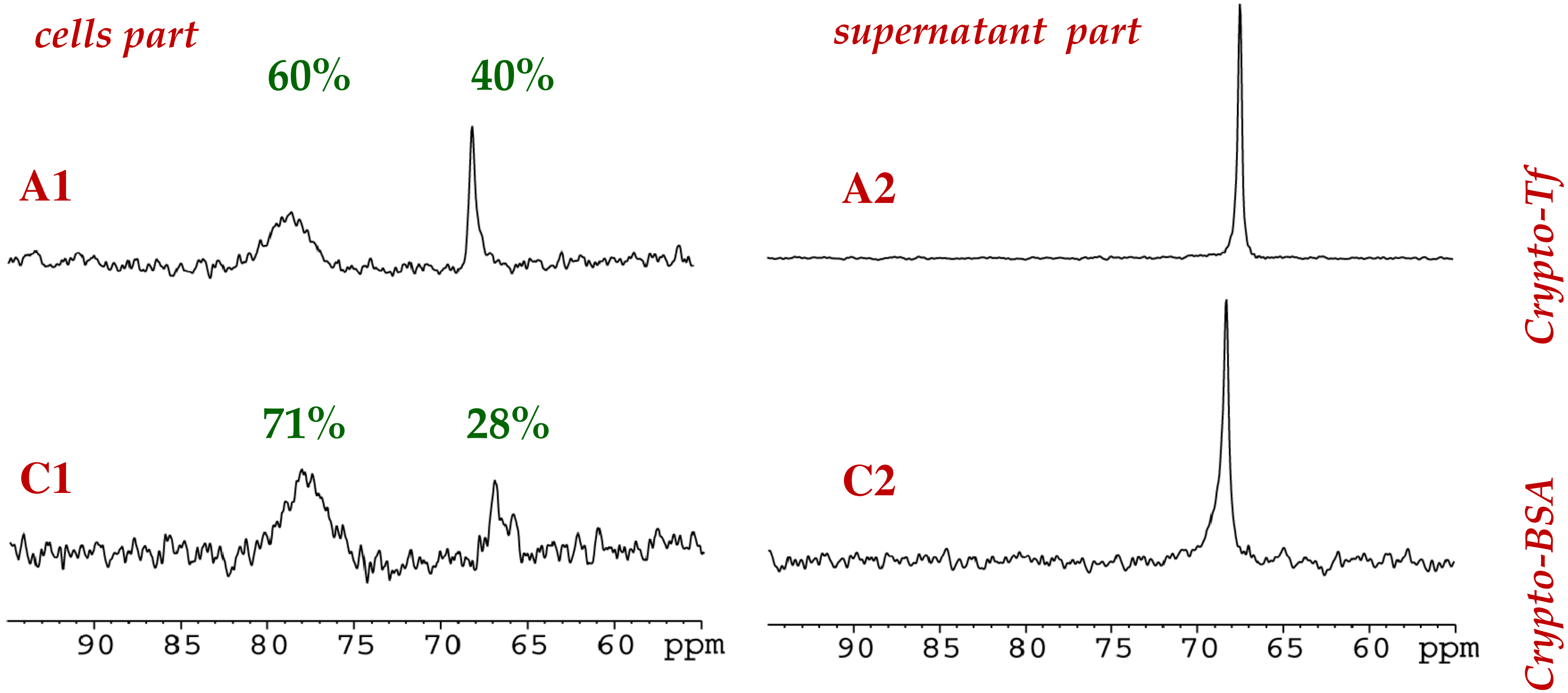


Cryptophane-
Rhodamine G-
BSA
R=2, R=2
37°C



Incubation 1h, concentration 200nM

Transferrin system:
^{129}Xe NMR results

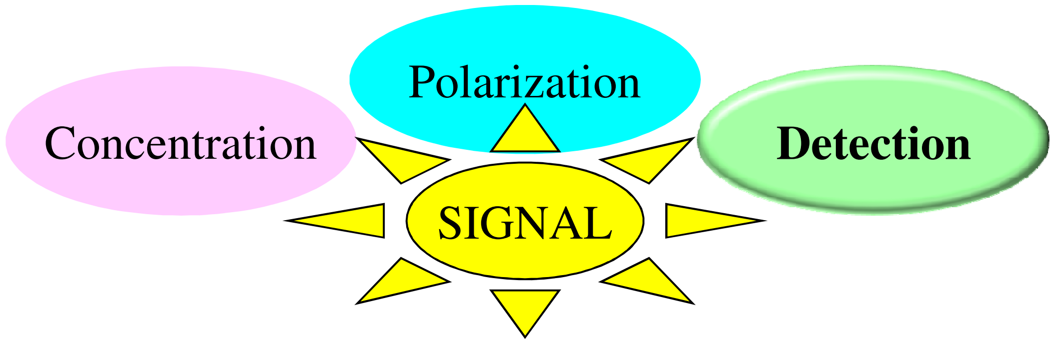


Conclusions on this study:

- Lack of specificity for the biosensor
- The cryptophanes may attract the biosensor in the cell membrane

- Despite the presence of Fe atoms, Xe relaxation is not too fast and allows detection of ~10^{13} sites
- First detection of an interaction with cells via ^{129}Xe NMR

Sensitivity of NMR



Alternative Detection Method: Spin Noise

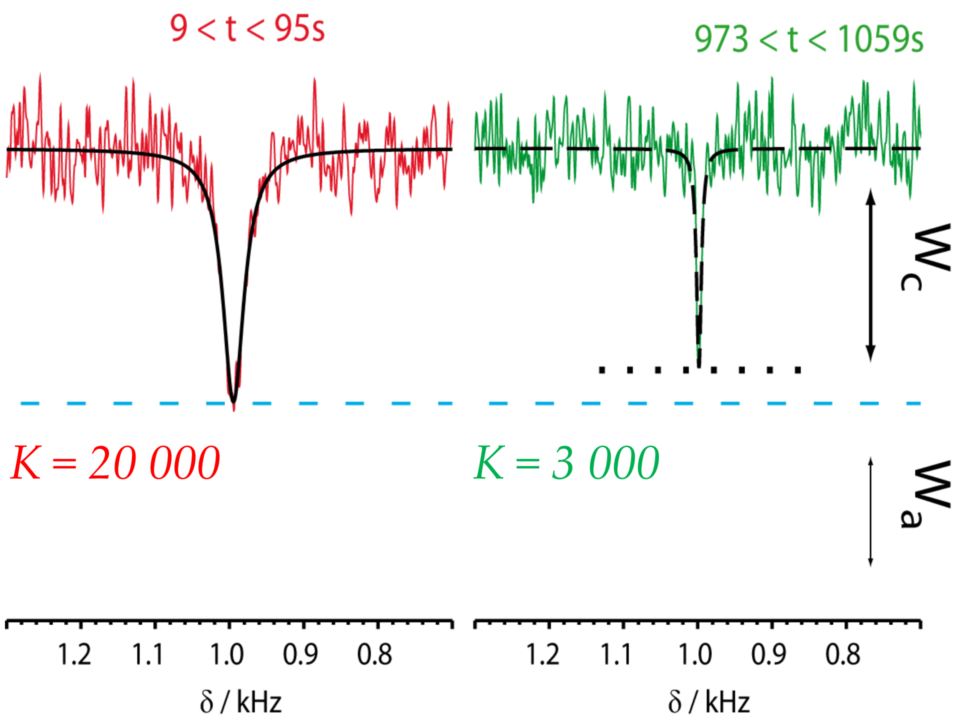
- No electromagnetic irradiation, just wait and record time-averaged power spectra
- Detection of the noise absorption by the spins at the Larmor frequency
- Does not depend on population differences, but phenomenon enhanced by radiation damping
- For a population of ***N*** non hyperpolarized spins, ratio of the signal obtained by spin noise over classical signal:

$$r = \frac{kT}{\mu B_0 \sqrt{N}}$$

Spin Noise Detection of Hyperpolarized Species

$$W^U(\omega) = W_c\left[1 + \frac{r_2^2 - (r_2 + K\lambda_r)^2}{(r_2 + K\lambda_r)^2 + (\omega - \omega_0)^2}\right] + W_a$$

K : polarization enhancement
λ_r: RD characteristic rate
r_2: transverse relaxation rate



Key condition:
$r_2 << K\lambda_r$

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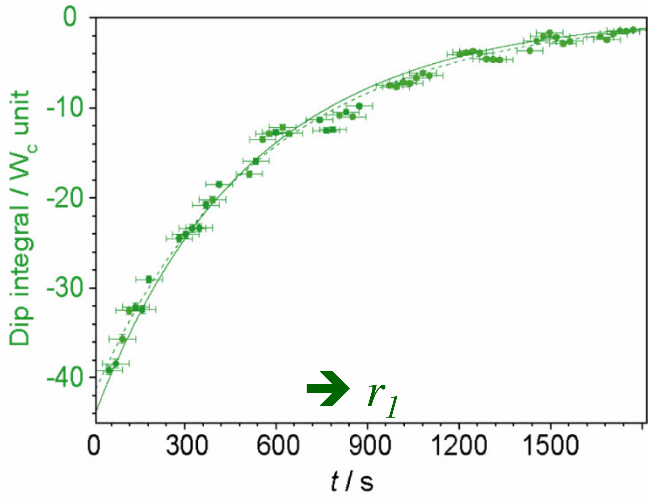
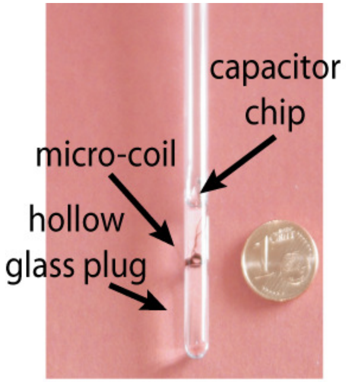
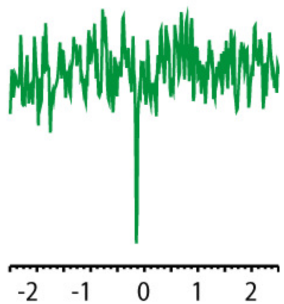
Spin Noise Detection of Hyperpolarized Species

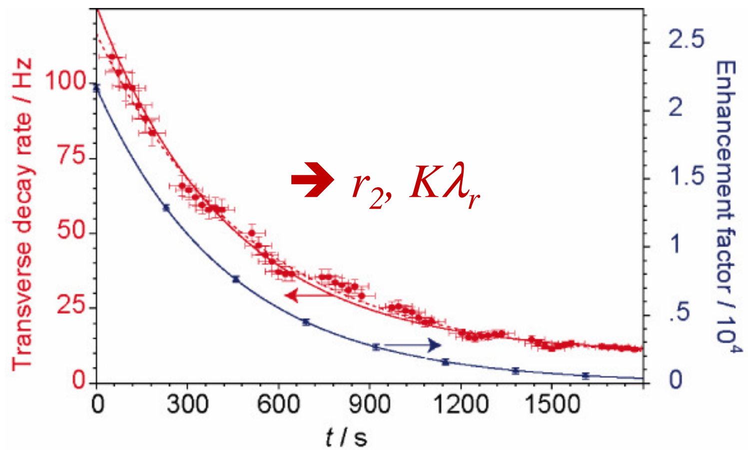
- No perturbation of the system
- Continuous monitoring of r_2, $K\lambda_r$, r_1
- No dependence on magnetic field
- Improvement possible:
  - probe Q and L values
  - preamplifier noise figure
- Detection of a small number of spins with micro-coils





Sensitivity improvements:
✓ ~ 4 for pulse
✓ ~ 50 for spin-noise





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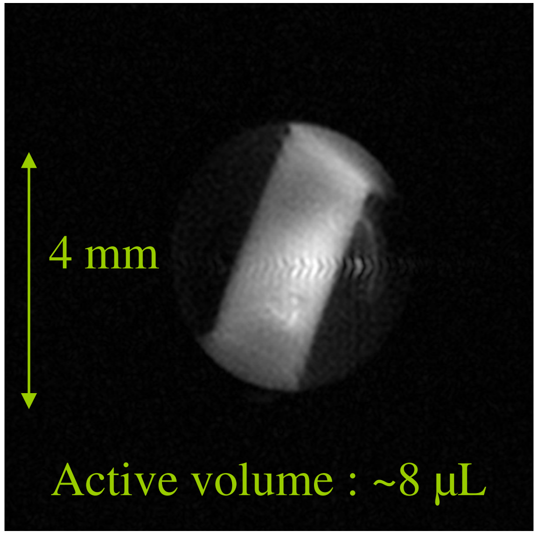
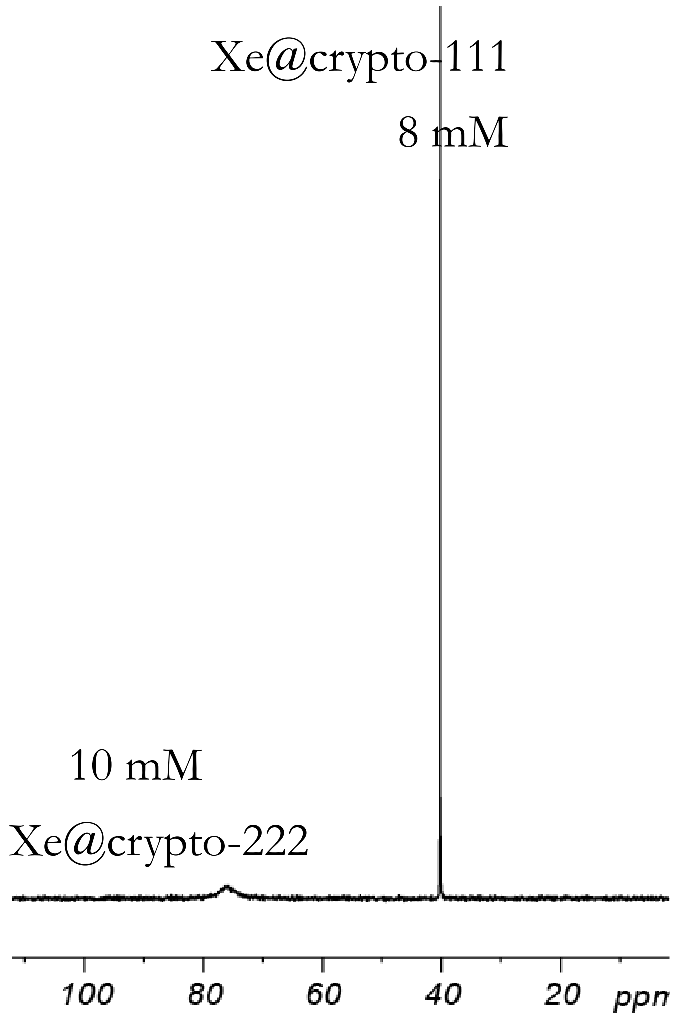
Perspectives

In vitro biosensing

- optimized xenon hosts
- activatable ^{129}Xe NMR-based biosensors
- use of inductively-coupled microcoils/spin noise

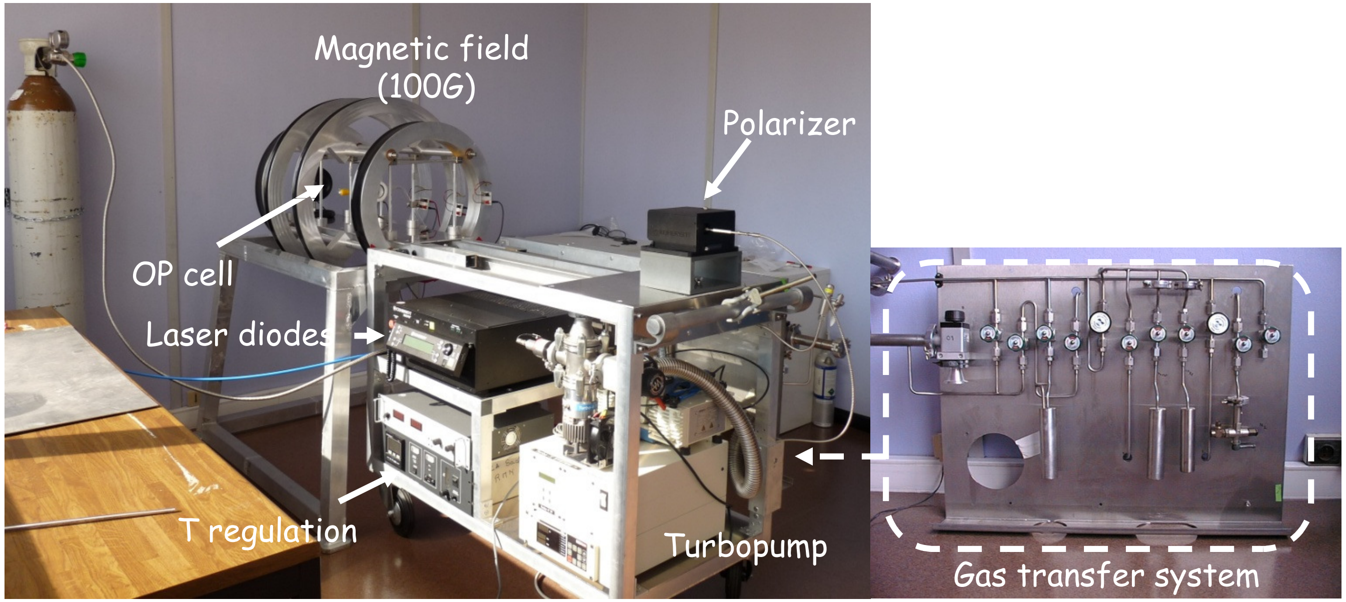




Perspectives

In vivo biosensing

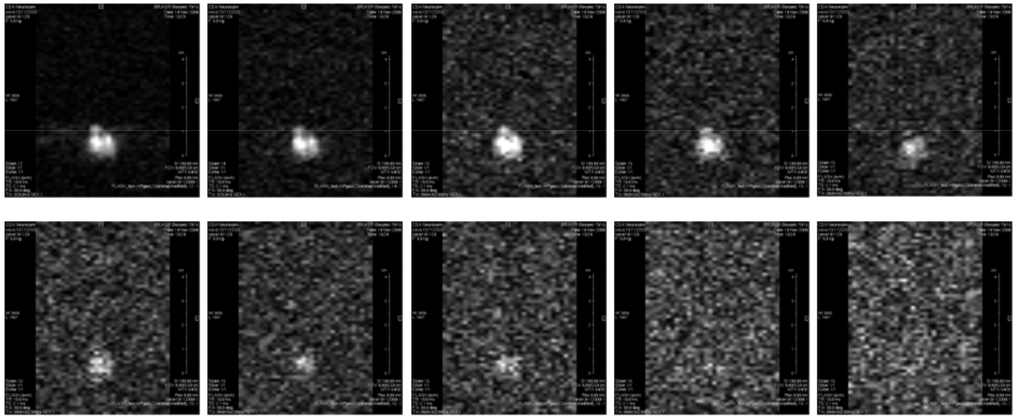
- New OP system for production of large quantities of polarized xenon

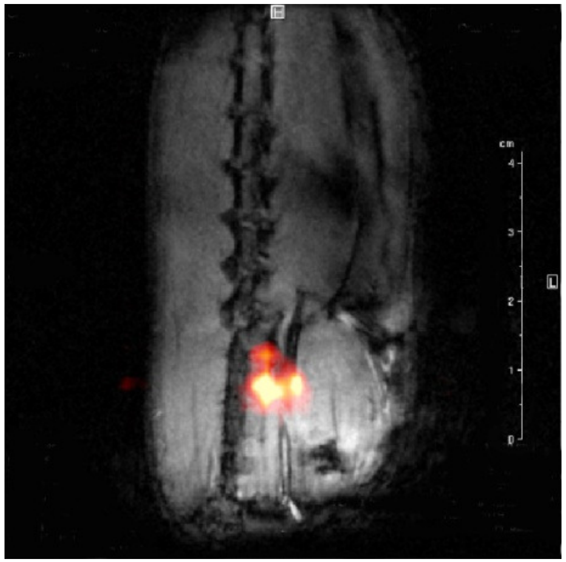


Perspectives

In vivo biosensing

- New OP system for production of large quantities of polarized xenon
- ^{129}Xe MRI experiments on rat:
  - injection
  - inhalation
- Fast MRI schemes:
  HYPERCEST-EPI, …



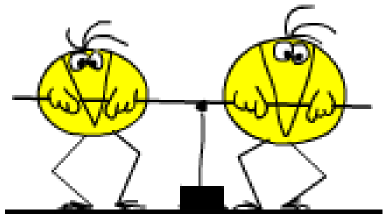


If successful, this imaging modality will render possible the monitoring of the evolution of targeted biological systems, through simple reintroduction of xenon after a first delivery of the biosensors.

Laser-Polarized xenon for NMR and MRI

Xenon Optical Pumping and NMR

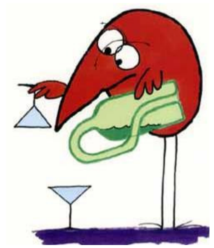
- Yves Boulard
- Hervé Desvaux
- Gaspard Huber
- Denis Marion
- Nawal Tassali



- Bruno Coltrinari *glassware*
- C. Chauvin, L. Liagre *workshop*

Xenon and cells, biological models

- Céline Boutin
- Marie Carrière
- Nadège Jamin
- François Leteurtre
- Alain Samson



MRI on rat

- Luisa Ciobanu
- Aude Padilla

Xenon hosts Chemistry

- T. Brotin
- J.-P. Dutasta
- H. Fogarty
- V. Roy
- A. Stopin

ECOLE NORMALE SUPERIEURE DE LYON

Agence Nationale de la Recherche GIP ANR