Applicazioni biomediche della Risonanza Magnetica e dei Materiali Magnetici

University researchers involved (NMR group, Dip. Fisica, UNIPV, http://arturo.unipv.it) :

Seniors : ***Borsa F., Carretta P., Corti M.,*** *Lascialfari A.* *(UNIMI),* ***Sanna S., Moscardini M. (Mariani M. – UNIBO, Rigamonti A.)***
collaboration with Nano R. (Biology) and Prof.s Altieri, Vidari (Chemistry), Dionigi (Medicine) groups

PhD and postdoc students : ***Bordonali L. , Filibian M., Orlando T., collaboration with Paolini A., Pasi F.***

UNIMI personnel: ***Orsini F., Arosio P.***

MAIN RESEARCH LINES

NOT EXHAUSTIVE

- ***Applications of magnetic nanoparticles to biomedicine – DIAGNOSIS AND THERAPY***
- ***Dynamic nuclear polarization – DIAGNOSIS (and THERAPY)***
- ***Magneto therapy – THERAPY (just begun)***
- ***Boron Neutron Capture Therapy – BNCT – THERAPY , see S. Bortolussi talk***

Suggested book : Magnetism in Medicine, eds. W. Andrä and H. Nowak, Wiley-VCH

Thanks are due to Q. Pankhurst and C. Sangregorio for contributions to slides

Laurea magistrale : courses directly involving NMR group

- ***Tecniche diagnostiche II*** ***(Carretta P., Lascialfari A.) : basics of NMR, MRI, Imaging***
- ***Strumentazione fisica biosanitaria*** ***(Corti M.) : MRI, ultrasounds, viscosity, CA, MFH***

Laurea triennale / laurea magistrale / PhD / postdoc / scuola specialità : Theses

Hospitals involved :

- **Pavia :** Policlinico S. Matteo, Ist. Neurologico "Mondino", Fondazione Maugeri
- **Milano :** Ospedale Niguarda, IEO, Ist. Mario Negri
- **contatti ulteriori :** Ist. Besta – Milano, Ist. Tumori – Milano, Policlinico - Milano

Companies :

Bracco SpA, Bruker Italia srl, Stelar srl

Students :

Laurea triennale : C. Cutaia, M. Mangiarotti, M. Mori, G. Savini, M. Montagna, A. Elia, T. Antonioli

Laurea magistrale : B. Comin, I. Zucca, M. Pasin, A. Vultaggio, M. Mangiarotti, F. Palesi, D. Panizza, F. Zucconi, A. Capozzi, T. Orlando, M. Alquati, L. Martignetti (UNITO), M. Nizzola (UNIMI)

PhD : F. Palesi, H. Amiri (UNIMI), K. Thangavel (UNIMI), L. Bordonali, T. Orlando

Postdoc : P. Arosio, M. Filibian, S. Velu, A. Gianella, A. Fiore, M. Marinone(…), M. Grandi (…), M. Mariani (…), F. Palesi, S. Pin, M. Bonora

Scuola specialità & companies : B. Comini, D. Panizza, F. Zucconi, F. Tedoldi, R. Melzi

Actual main collaborations
(magnetism, MRI, synthesis, hyperthermia)

- Dipartimento di Chimica, Università di Roma, gruppo prof. G. Ortaggi
- Dipartimento di Chimica, Università di Cagliari, Dr. M.F. Casula
- Dipartimento di Chimica, Universita' di Modena, gruppo prof. A. Cornia
- Dipartimento di Chimica, Universita' di Firenze, gruppo prof. D.Gatteschi
- Dipartimento di Chimica Fisica, Universita' di Pavia, gruppo prof. P. Ghigna
- Dipartimento di Chimica, Università di Pisa, Prof. E. Chiellini
- Dipartimento di Chimica, Università di Bologna, Prof. M. Comes Franchini
- Dipartimento di Chimica, Università degli studi di Milano, prof. P. Ferruti e E. Ranucci
- Dipartimento di Chimica, Università degli studi di Milano, prof. G. D'Alfonso
- Dipartimento di Scienze Chimiche, Università degli studi di Padova, Dr. V. Amendola
- Dipartimento di Scienze Chimiche, Università degli studi di Catania, Prof. G. Vecchio
- Dipartimento di Fisica, Università di Milano Bicocca, gruppo prof. C. Riccardi
- Dipartimento di biologia e biotecnologie "L. Spallanzani", Università degli studi di Pavia, Prof. R. Nano
- Dipartimento di Chimica, Università di Pavia, Prof. Vidari
- Facoltà di Medicina, Università di Pavia, Prof. Dionigi
- Dipartimento di Fisica, Università di Pavia, Prof. Altieri
- Dipartimento di Scienze Farmacologiche, Universita' di Milano, Prof. R. Paoletti, Prof. E.Tremoli, Dr.U.Guerrini, Dr.G.Sironi
- Dipartimento di Scienze Morfologiche-Biomediche, Università degli studi di Verona, Prof. P. Marzola, Prof. A. Sbarbati
- Dipartimento di Scienze Farmacologiche e Biomolecolari, Università degli studi di Milano, Prof.ssa V.F. Sacchi, Dr.ssa A. Rizzo
- Dipartimento di Chimica, Università degli studi di Milano, Dr. M. Scavini
- INFM-CNR,National Nanotechnology Laboratory, Dr. T. Pellegrino, Dr. D. Cozzoli, Dr. L. Manna, Prof. R. Cingolani
- Department of Chemistry, Humboldt Universitat – Berlin (Germania), Prof. N. Pinna

Regional Center of Advanced Technology and Materials, Olomouc (Repubblica Ceca), Dr. G. Zoppellaro

•FORTH (Foundation for Research and Technology – Heraklion, Greece), Prof. A. Lappas

•Dept. Chemistry, Università di Bordeaux, Prof. S. Lecommandoux

•Dept. Of Physics, University of Zaragoza (Spagna), Prof. F. Palacio e Dr. A. Millan

•Departamento de Química Inorgánica, Universidad de Granada (Spagna), Dr. J.M. Dominguez-Vera

•CNRS and University of Montpellier (Francia), Dr. J. Larionova, Dr. Y. Guari

•Pasteur Institute of Tehran (Iran), Dr. M. Mahmoudi

•Phillips Marburg University (Germany), Dr. W. J. Parak

•Policlinico S. Matteo, Pavia, Dr. R. Di Liberto (direttore Fisica sanitaria)

•Istituto Neurologico "Mondino", Pavia, Prof. E. D'Angelo (anche Università degli studi di Pavia) e Dr.ssa F. Palesi

•Ospedale Niguarda, Milano, Dr. A. Torresin (direttore Fisica Sanitaria)

•Istituto Europeo di Oncologia (IEO), Milano, Dr.ssa D. Origgi

•Bracco SpA, Milano (Italia), dr. V. Lorusso

•Centro Ricerche Colorobbia, Vinci (FI) (Italia), Dr. G. Baldi, Dr.ssa L. Niccolai

•Stelar srl, Mede (PV) – Italia, Ing. G. Ferrante

Projects open

- FIRB Riname – PI prof. D. Gatteschi (INSTM – FI)
- Fondazione Cariplo “Chemical synthesis and characterization of magneto-plasmonic nano-heterostructures” – PI Prof. P. Ghigna (INSTM-PV)
- AIRC “Magnetosomes as nanotechnology platform for thermotherapy of tumour”, PI A. Sbarbati (UNIVR)
- European project NANOTHER, PI Gaiker (Spain), **just ended**
- INSTM – Regione Lombardia MAGNANO, PI A. Lascialfari
- Contract UNIPV – Bracco SpA (DNP)

Experimental techniques at NMR group

Universita’ degli studi di Pavia

- **MRI Esaote , Artoscan**, H= 0.2 Tesla
- **NMR : broadband spectrometers** (Bruker, Tecmag, Mid-Continent) covering a frequency range 4-400 MHz. Magnetic field 0-9 Tesla. Temperature : 0.3-1000 K.
- **Quantum Design SQUID magnetometer**. Temperature: 1.7-800K.

  Magnetic Field : -7 ÷ 7 Tesla.
- **Adiabatic calorimeter,** 1.5 < T < 300 K
- **”Wide-band” EPR**

Universita’ degli studi di Milano

- **Atomic Force Microscopy** / Scanning Tunneling Microscopy / Magnetic Force Microscopy - Autoprobe CP Research System - Veeco. Working temperature range 0-60°C.
- **NMR : broadband spectrometer** Stelar Spinmaster. 5-70 MHz. Temperature : 4.2-350 K. Magnetic field 0-1.4 Tesla.
- **NMR relaxometer**, 10 KHz < f < 10 MHz. Temperature 150 < T < 350K

  (**MRI : Bruker AMX200**, 4.7 Tesla, super-wide-bore, rats and mice coils. Physiological parameters’ control)

Available equipments : broad-band NMR, magnetometry and relaxometry

SMART Tracer
Frequency : 10KHz – 10MHz

FT-spectrometers and electromagnets

9 Tesla magnet for broad-band NMR

Esaote Artoscan 0.2 Tesla MRI Imager

SQUID magnetometer

Cryogenics

Clinical techniques related to our research

Magnetic Fluid Hyperthermia (MFH) or Magnetothermia

TUMOUR THERAPY

- **Heating** through application of **AC magnetic field** via activation of 12 nm amino-silane coated Fe_3O_4 MNP directly implanted in the tumour (**glioblastoma**) mass at high doses (ca. 50 mg/cm^3)
- Typically : $\nu \sim 100$ kHz, amplitude 10 kA/m
- Minor side effects

- Typical values of the reported **specific loss of power, SLP or SAR** (the energy converted into heat per mass unit) are : 10÷200 W/g
- Exceptions :
  - 35 nm bacterial magnetosomes (960 W/g at 410 KHz and 10 kA/m)
  - 16 nm γ-Fe_2O_3 N P (1650 W/g at 700 kHz and 24.8 kA/m, 300 W/g at 11 kA/m)

Clinical techniques related to our research

Typical MRI apparatus for clinical use, magnetic field H = 1.5 Tesla

Magnetic Resonance Imaging (MRI) DIAGNOSIS

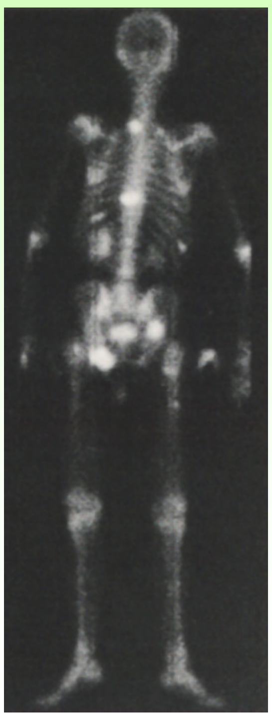
MRI Timeline

1937 MR phenomenon - Rabi
1946 MR phenomenon - Bloch & Purcell
1952 Nobel Prize - Bloch & Purcell
1950-70 NMR developed as analytical tool
1972 Computerized Tomography
1973 Backprojection MRI - Lauterbur
1975 Fourier Imaging - Ernst
1977 Echo-planar imaging - Mansfield
1980 FT MRI demonstrated - Edelstein
1986 Gradient Echo Imaging - NMR Microscope
1987 MR Angiography - Dumoulin
1991 Nobel Prize - Ernst
1992 Functional MRI
1994 Hyperpolarized ^{129}Xe Imaging
2003 Nobel Prize - Lauterbur & Mansfield

Why MRI?

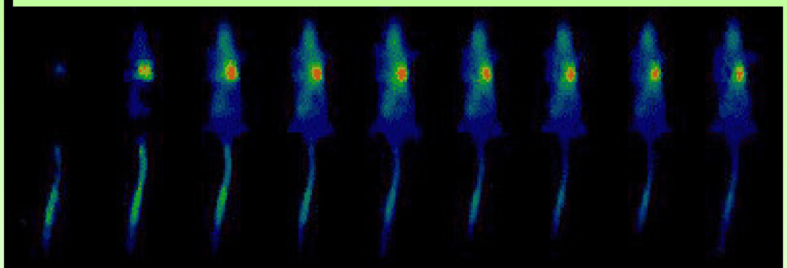
Nuclear Medicine:

- Poor spatial resolution
- Poor temporal resolution
- High sensitivity
- Reporters: radionuclides



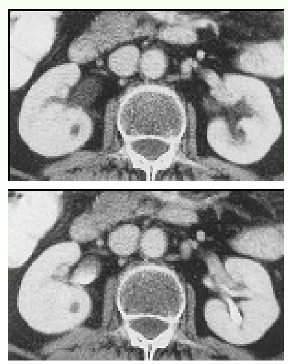
Optical Imaging:

- Poor spatial resolution
- Poor temporal resolution
- high sensitivity
- Reporters: luminescent probes



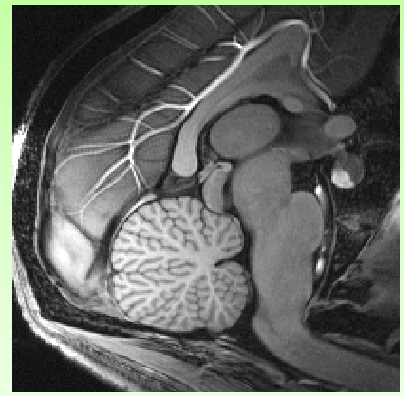
X-Ray (CT):

- Good spatial resolution
- Good temporal resolution
- Low sensitivity



MRI:

- **Non-invasive**
- Good spatial resolution
- Good temporal resolution
- Low sensitivity

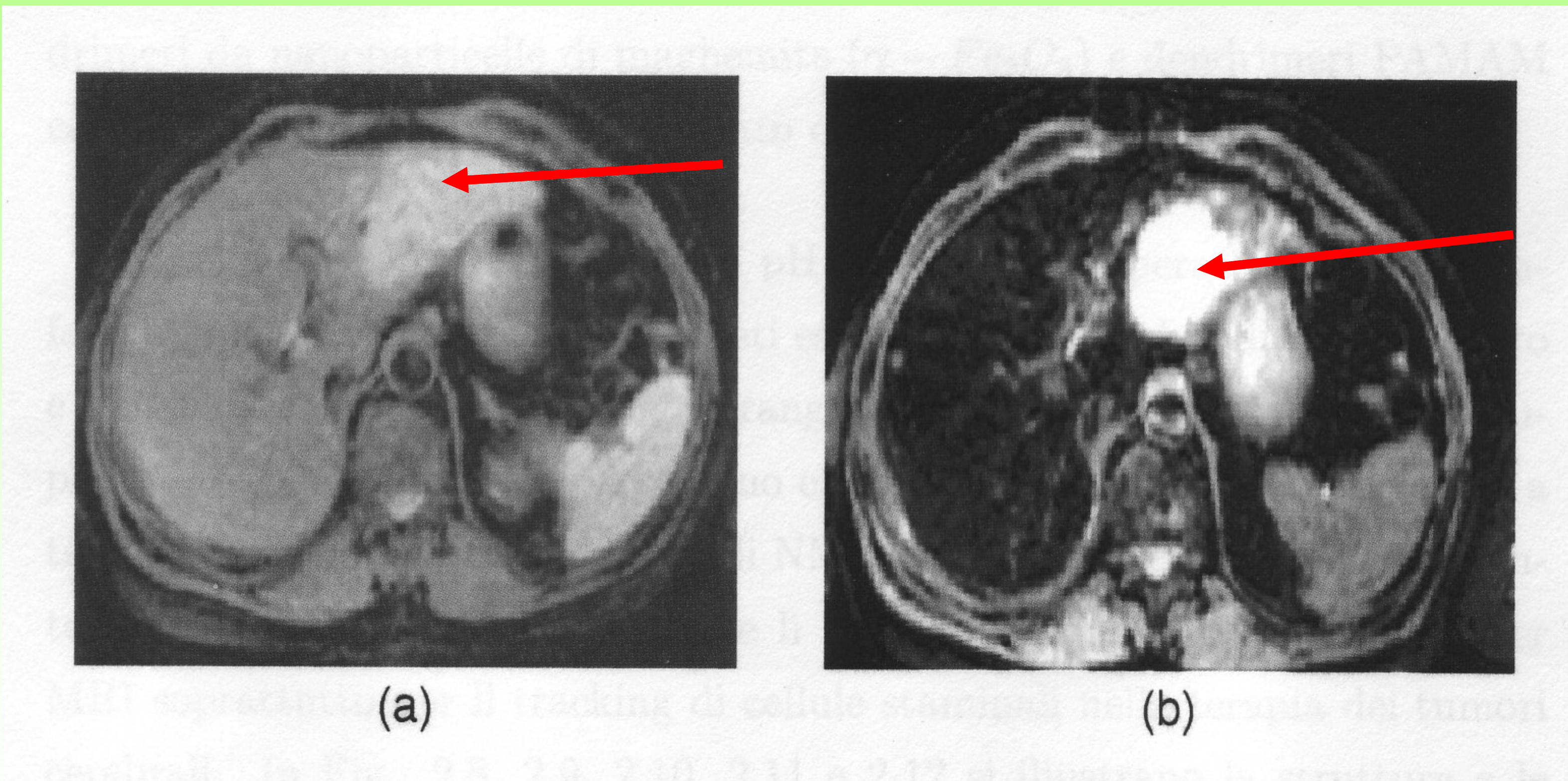


Why MRI contrast agents ?

Liver tumour detection by "negative" SP-CA

- Generally the **negative CA** are based on **superparamagnetic nanoparticles**

Example : liver tumour



without CA **with CA**

Research interest in magnetic materials : different mechanisms

Sensing
(MRI, Sentimag, MEG-SQUID,…)

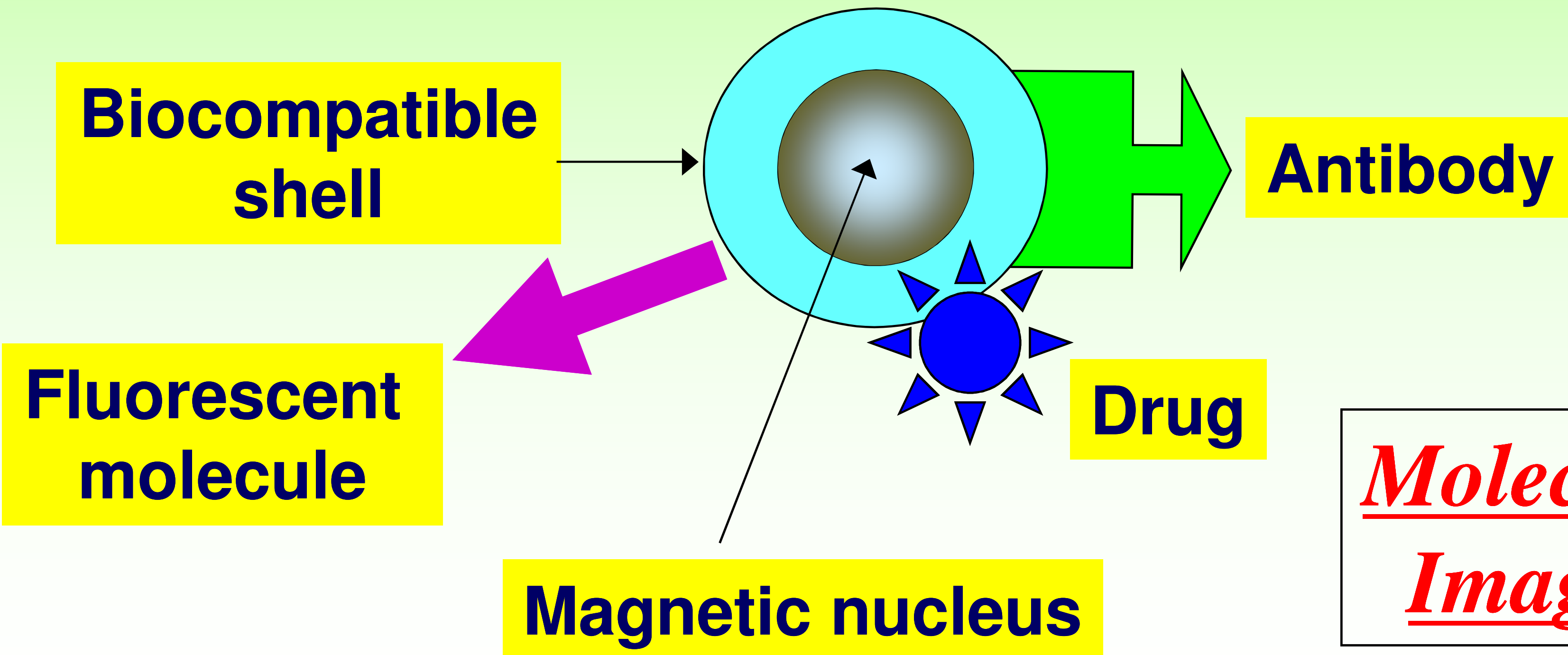
Moving
(navigation)

Heating
(Magnetic Hyperthermia)

Ideal magnetic nanoparticle

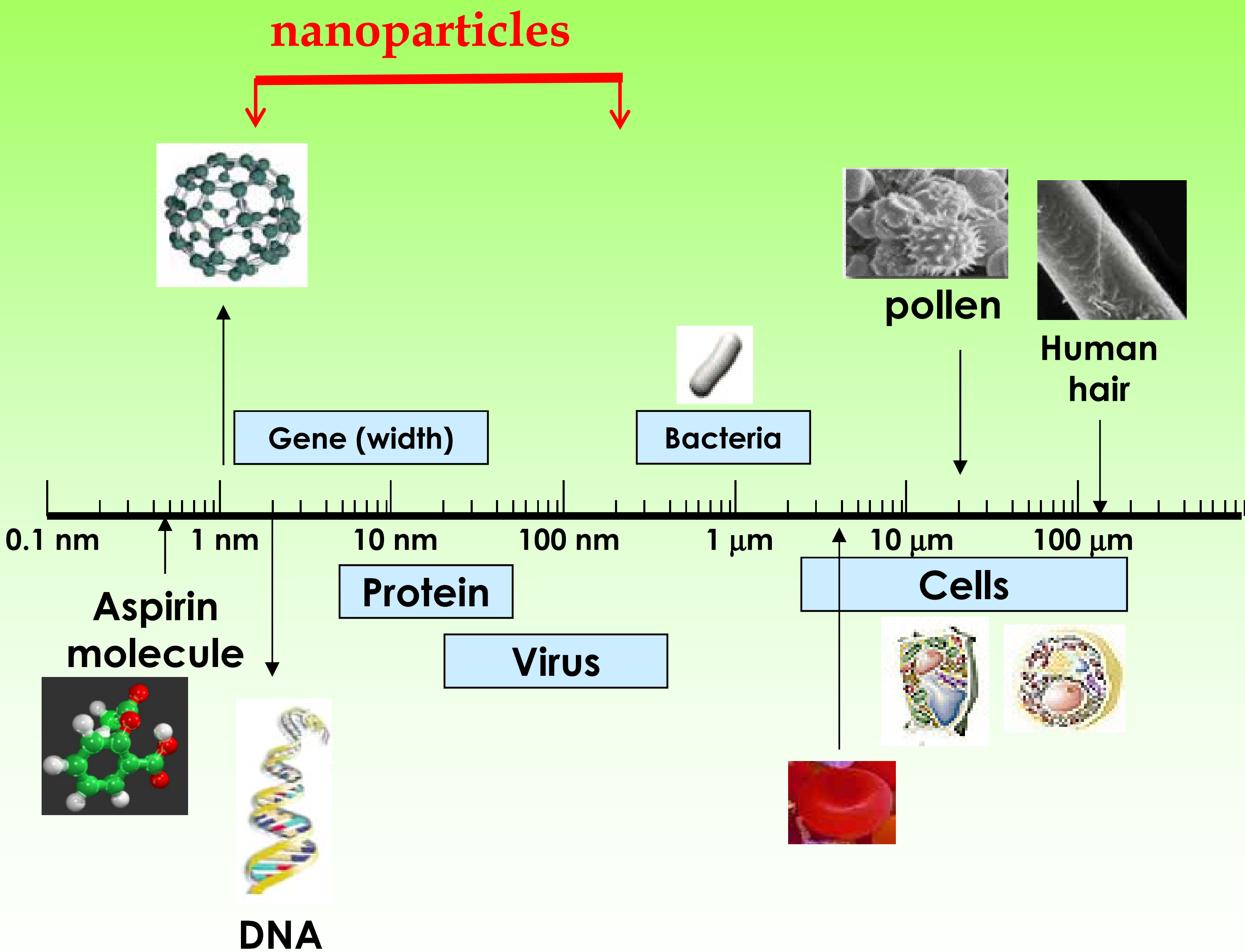
Magnetic Nanoparticles in <u>Theranostics</u>:

Diagnostics : **<u>MRI</u>, sensing, fluorescence**
Therapy : **magnetothermia, drug delivery**



<u>Molecular Imaging</u>

Typical dimensions in nanomedicine



MNPs targeting (in vivo) & uptake

After MRI contrast agents injection

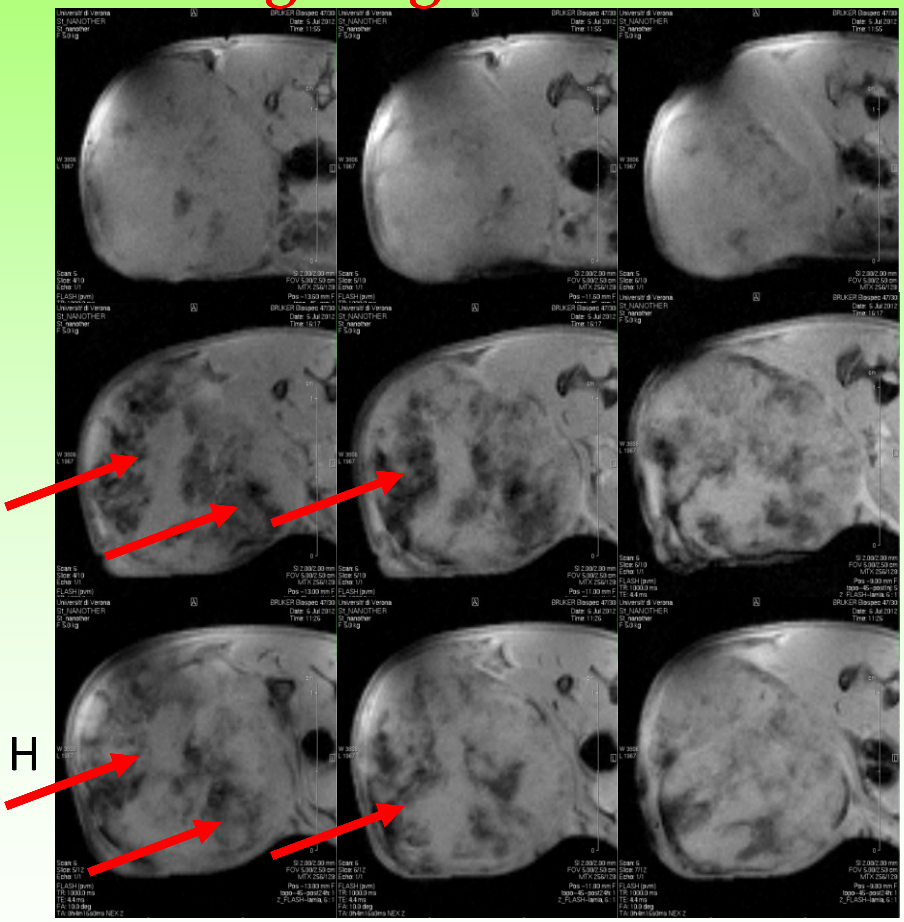
NP- Pisa- folic acid (Gradient-echo)

Targeting effective

PRE

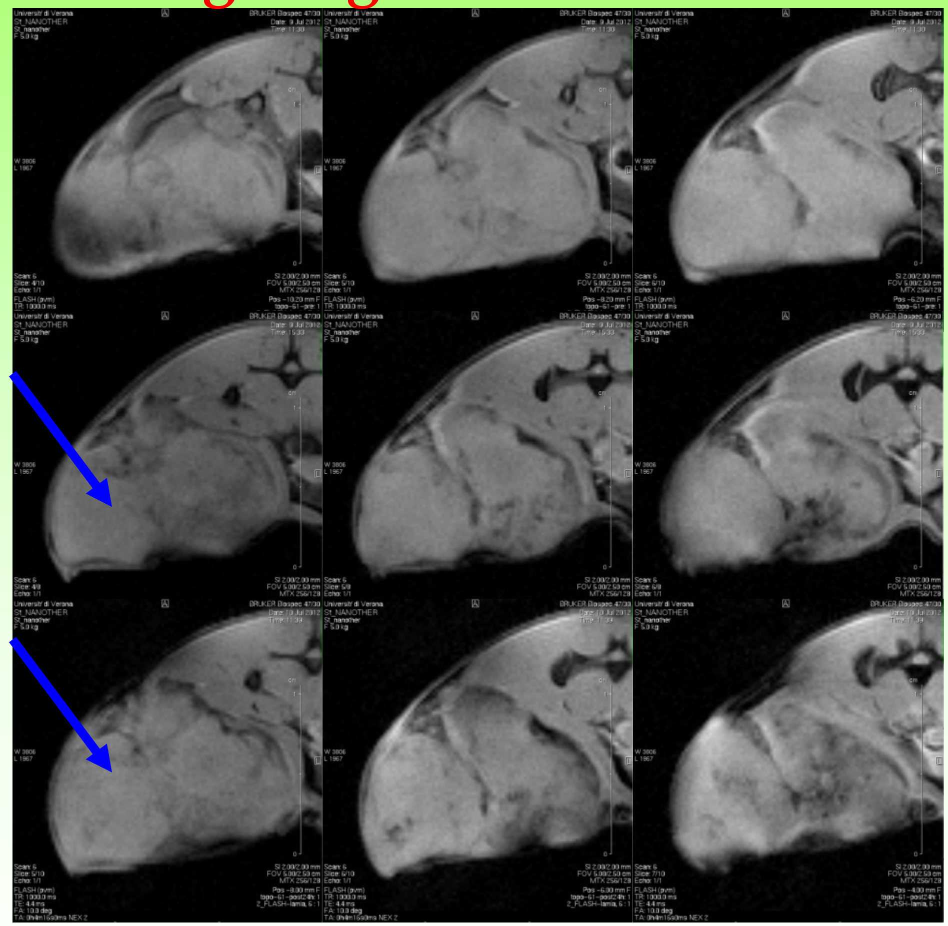
POST

POST 24 H



NP- Pisa- without folic acid (Gradient-echo)

Targeting NOT effective

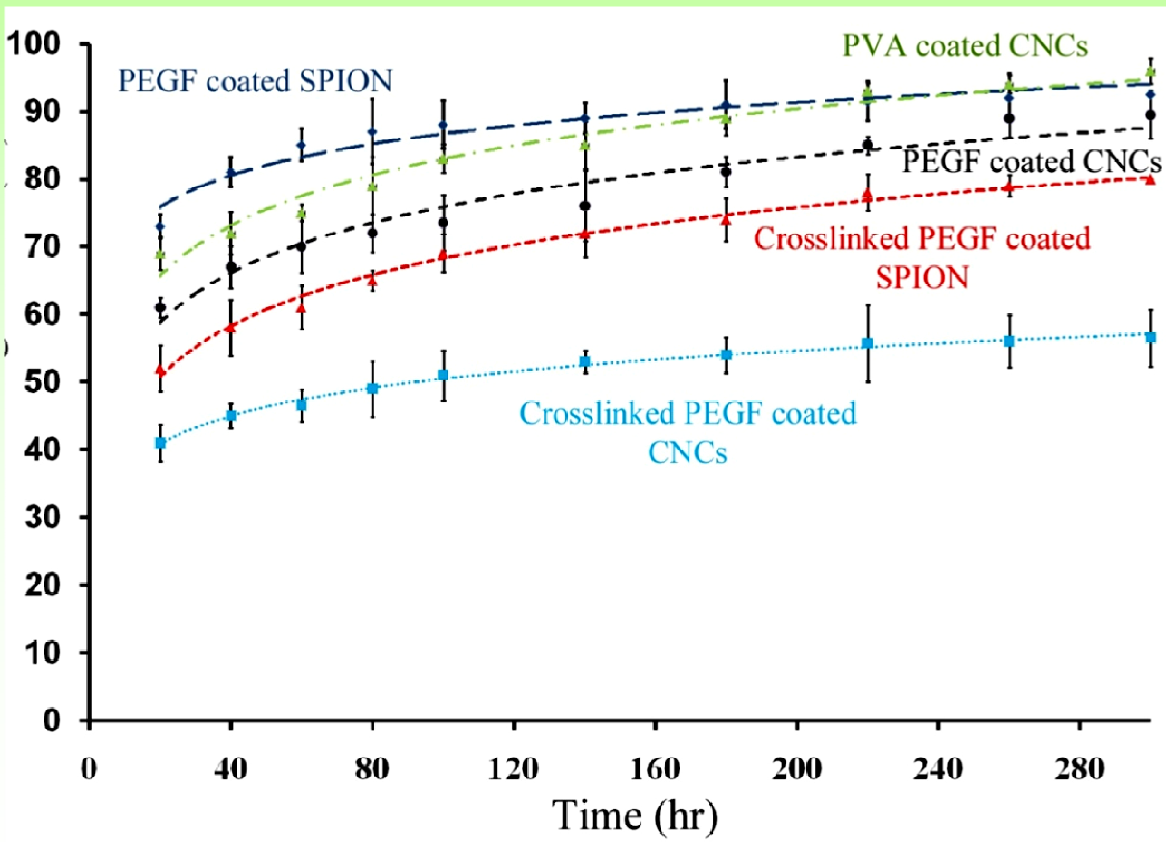


- **MRI images of animal model with breast tumour**
- **Uptake studies in progress (R. Nano)**

Therapy by novel MNPs : drug release

Percentage release of Tamoxifen over 12 days (in vitro)

Sharif University of Technology.



Examples of biosensors

First Biosensor ! → *Coal miners' biosensor*

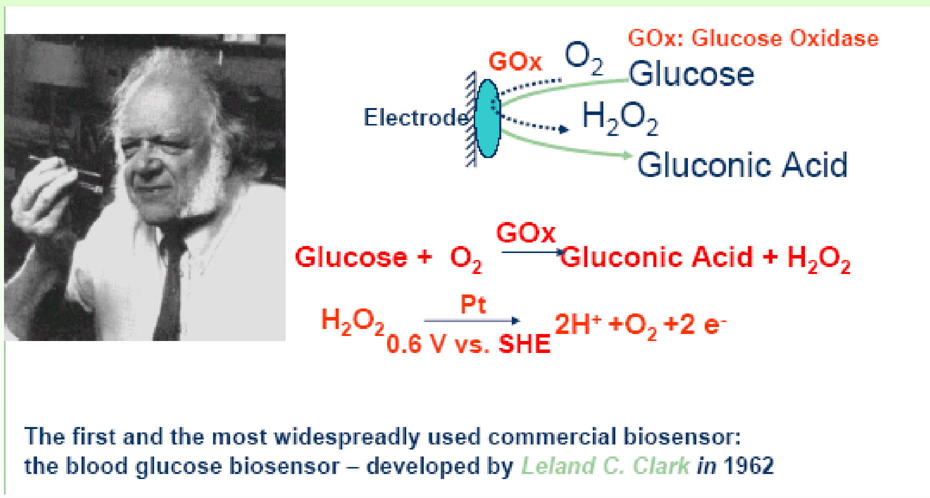
Commercial Biosensors

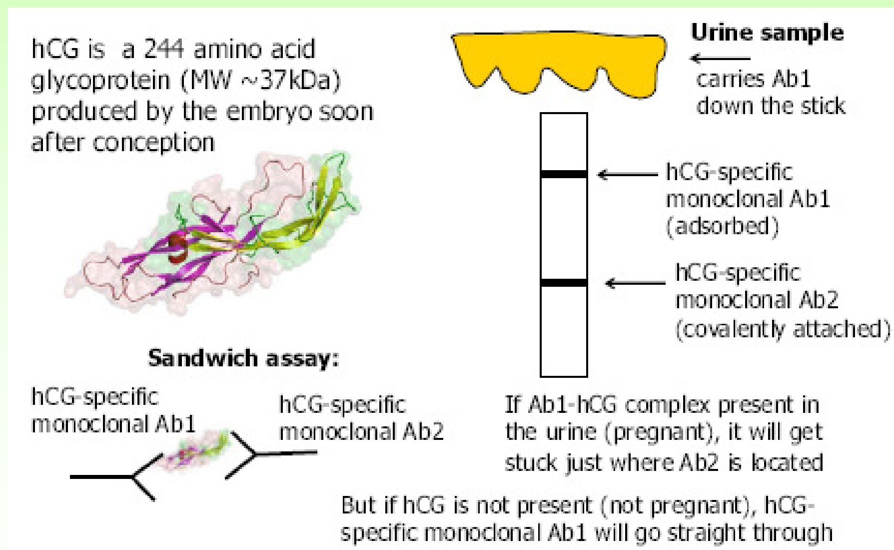
Glucose biosensor

By Johnson & Johnson

Pregnancy test

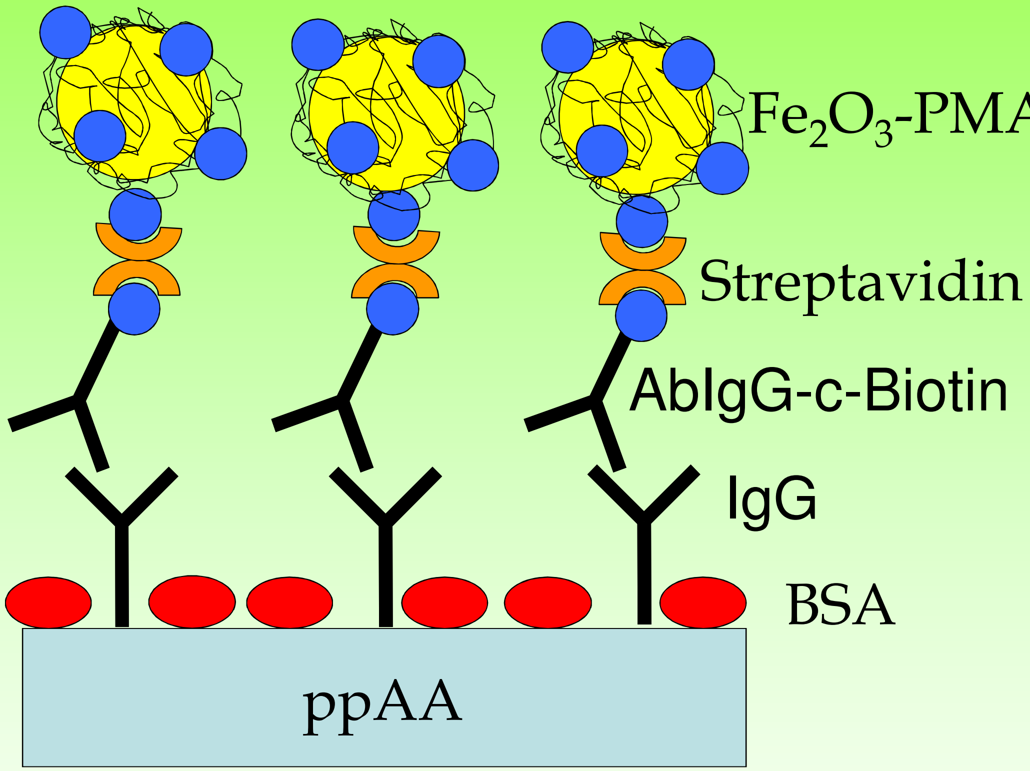
By Gima





Solid magnetic biosensors

Surface modification approach



- Plasma Deposited Poly Acrylic Acid (ppAA) [*]
- Adsorption of human IgG
- Blocking of the unreacted surface groups by BSA
- Reaction with biotinylated Ab-IgG molecules at different concentrations
- Absorption of streptavidin
- Absorption of *biotinylated modified γ-Fe_2O_3 superparamagnetic nanoparticles*

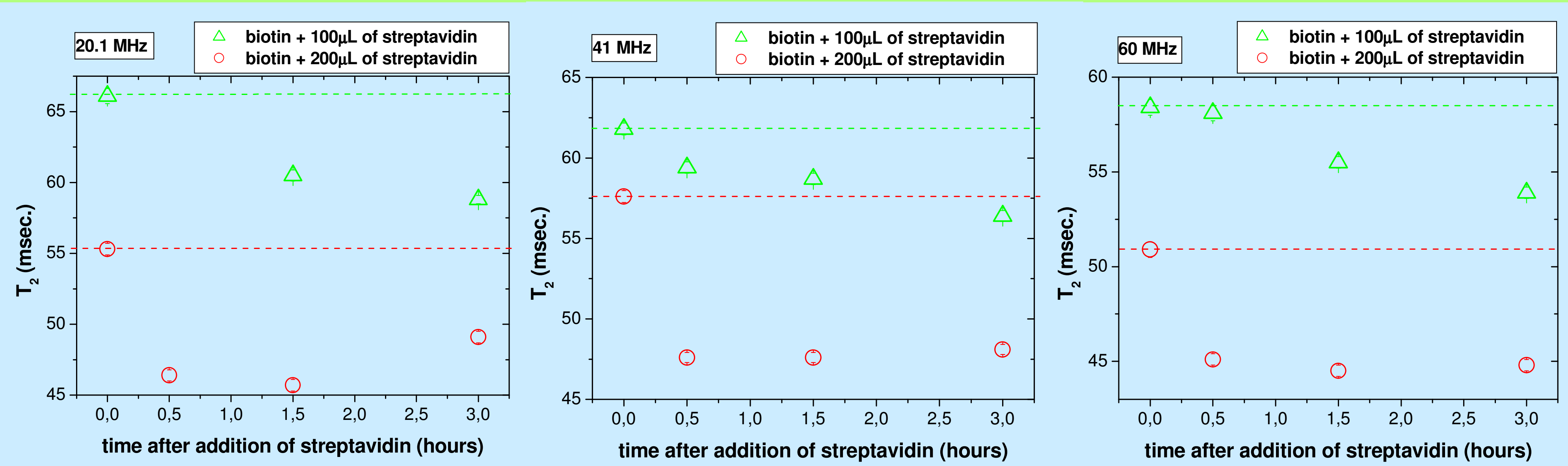
[*] F. Bretagnol, A. Valsesia, G. Ceccone, P. Colpo, D. Gilliland, L. Ceriotti, M. Hasiwa, and F. Rossi *Plasma Processes and Polymers 3*, 443 (2006).

Solid magnetic biosensors

Results of the approach

[1]H-NMR measurements

* Room temperature * [1]H-NMR relaxation times T_1 and T_2 evaluated at 20.1, 41 and 60 MHz



as a function of time after the addition of streptavidin and quantity of streptavidin ([Strept] = 1mg/mL)

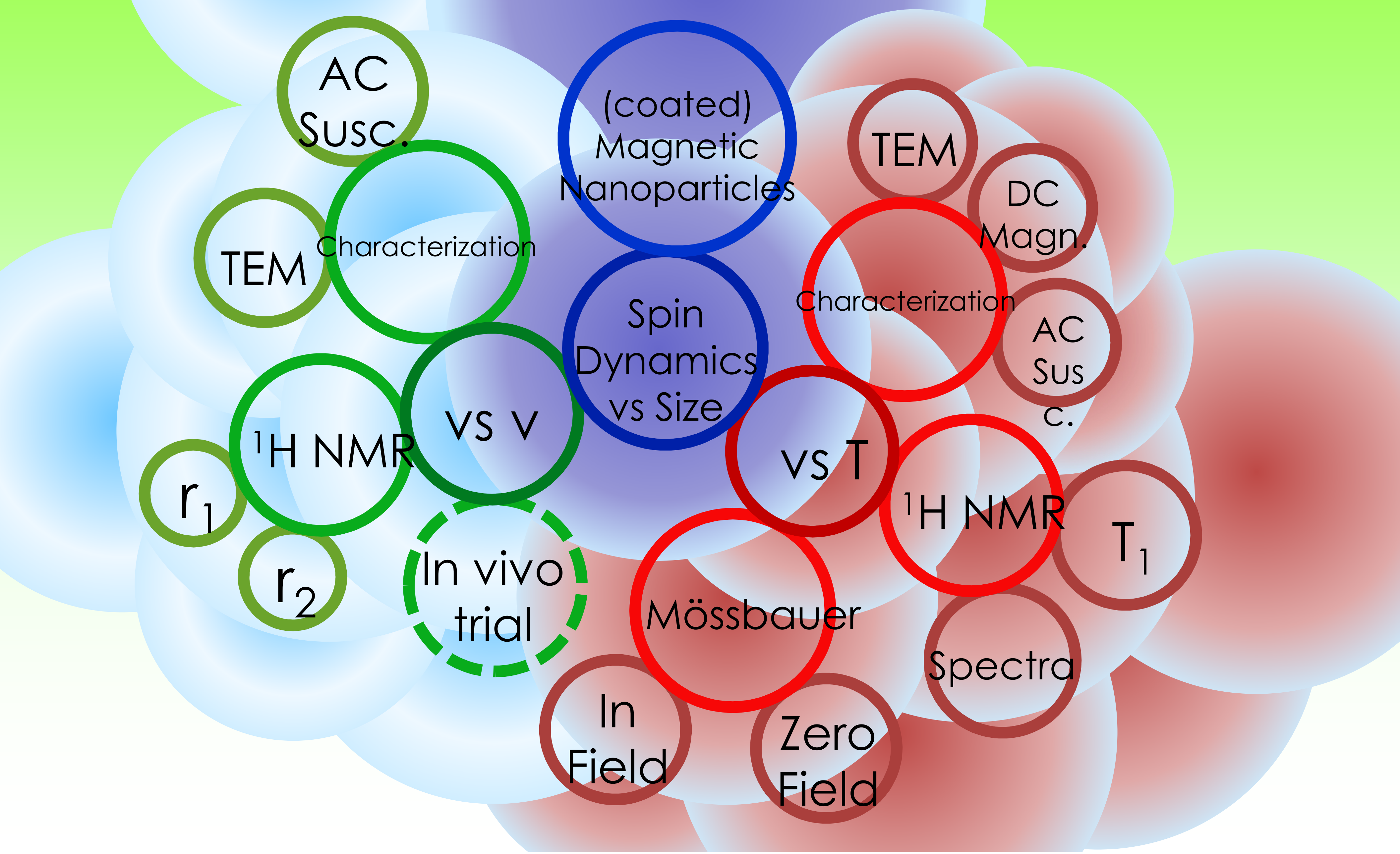
The longitudinal relaxation time T_1 not reported ⇒ under study

A 10-15% change of the transverse relaxation time T_2 ⇒ <u>Sensitivity of NMR</u>

! Detection of probe-analyte interaction ! … Clusters !

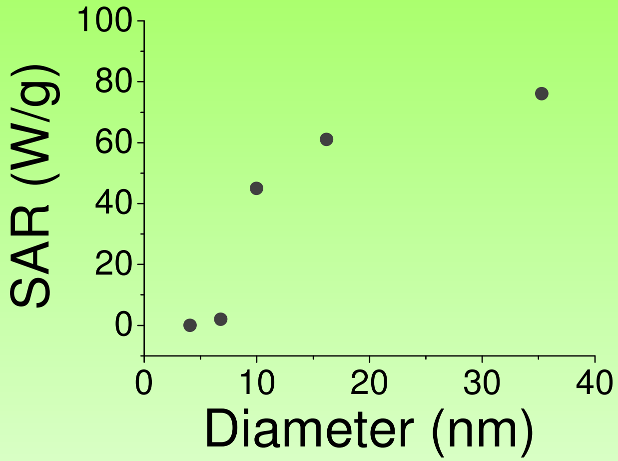
! Analyte not modified !

MNPs : from fundamental properties to applications

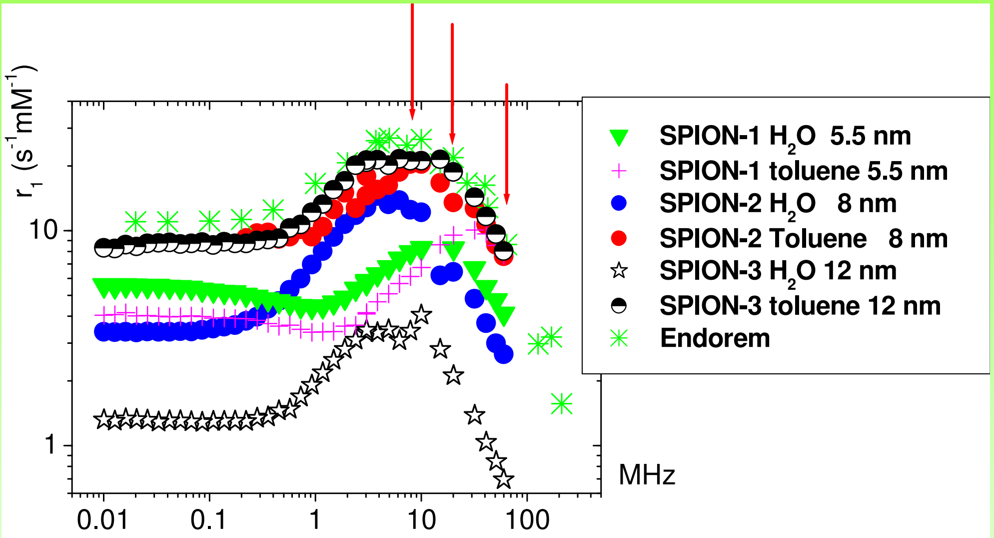


MNPs : from basic properties to applications

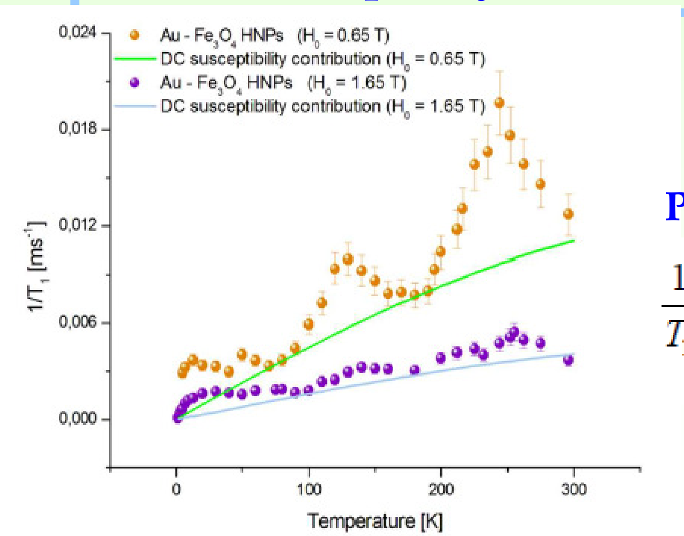
SAR in Magnetic Fluid Hyperthermia : Study of influence of microscopic features



NMR relaxometry at room temperature



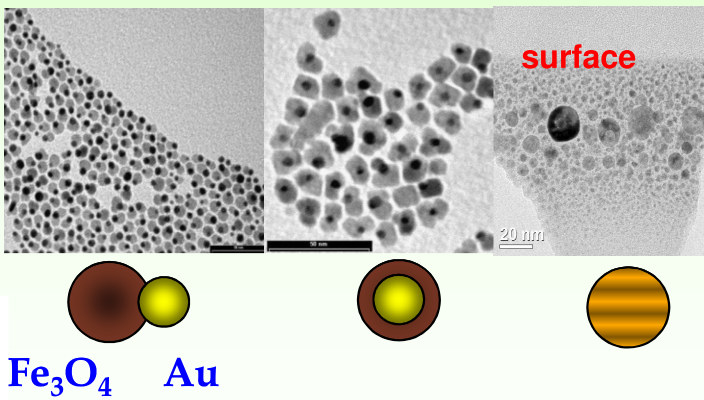
Magneto-optical nano-devices : ferrites+gold plasmon resonance and spin dynamics



Physical model for spin dynamics :

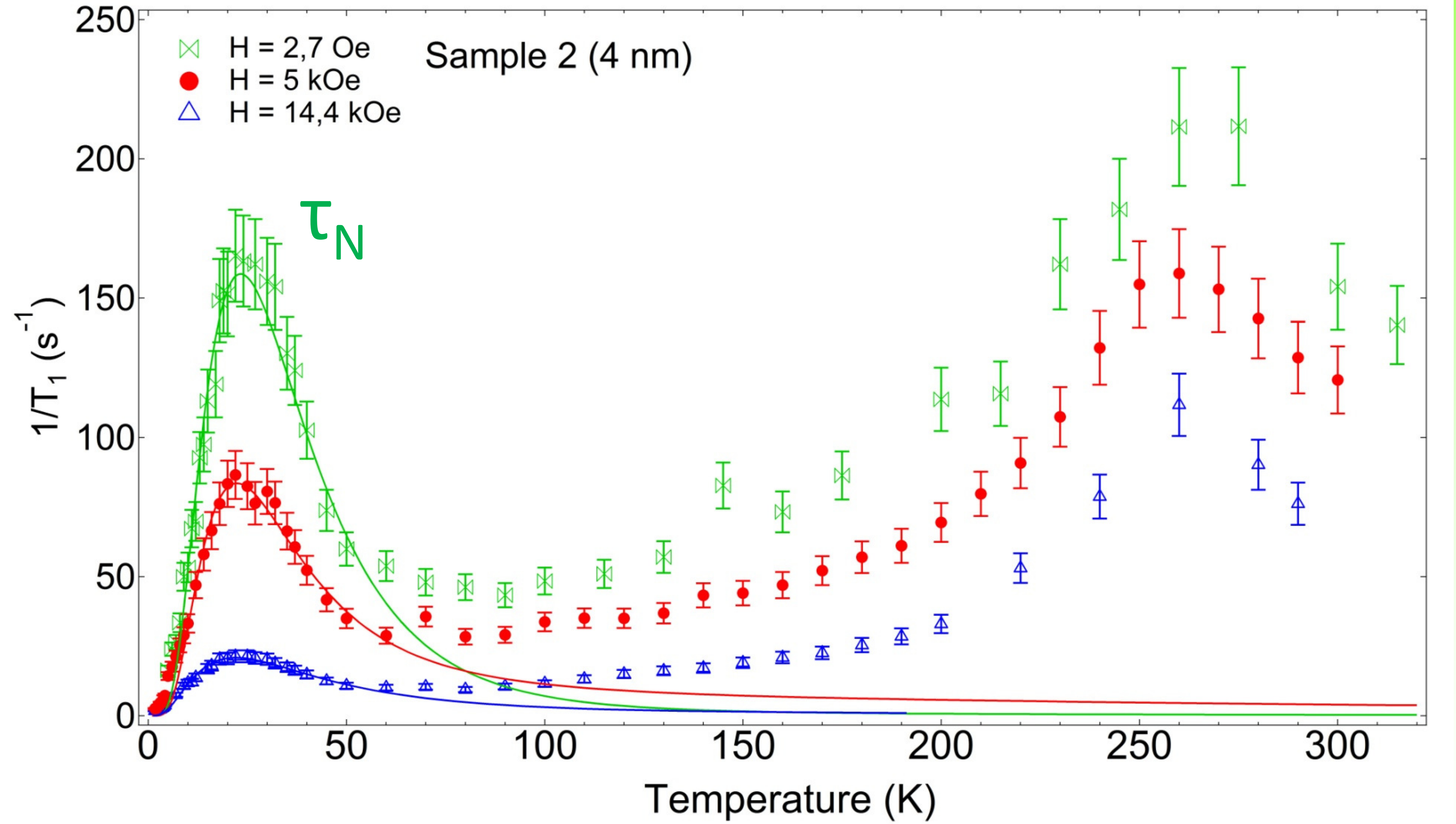
$$\frac{1}{T_1} = A\chi T\left(\frac{\tau_R}{1+\omega_L^2\tau_R} + \frac{\tau_N}{1+\omega_L^2\tau_N} + \frac{\tau_?}{1+\omega_L^2\tau_?}\right)$$

Magneto-plasmonics



MNPs : fundamental magnetic properties

NMR vs temperature : disclosing spin dynamics & fundamental magnetism



Dynamic Nuclear Polarization : a novel contrasting technique and its principles

- Transfer of polarization from the electron to the nuclear spin system under irradiation of the electronic resonance

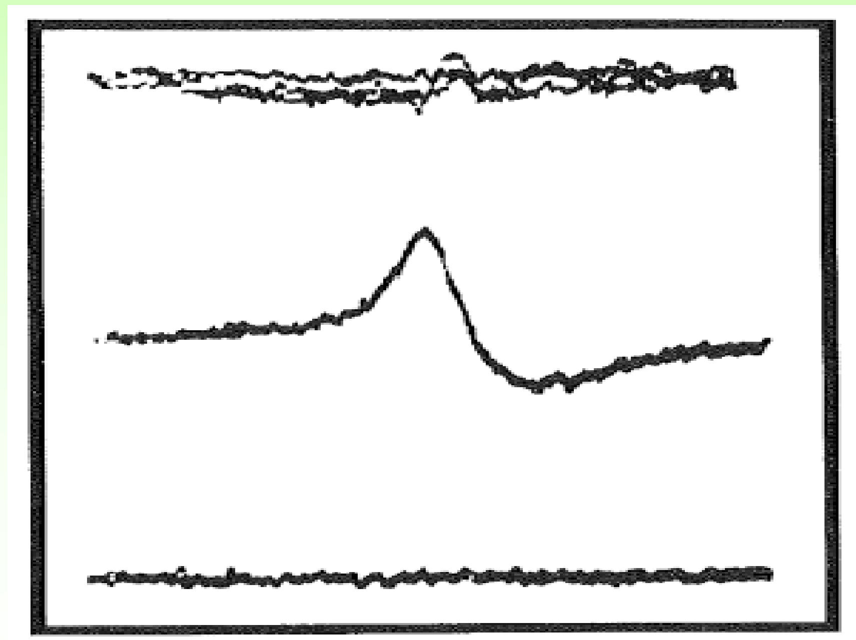
$$P_n(\%) = \varepsilon\, P_n^{thermal} = \varepsilon \tanh\left(\frac{\gamma \hbar H_0}{2K_B T}\right) \propto \frac{\varepsilon}{T}$$

<u>NON EQUILIBRIUM HYPERPOLARIZED STATE</u>

- In Metals and solutions
  Overhauser Effect

- In solids doped with radicals
  Solid State Effect (SE)

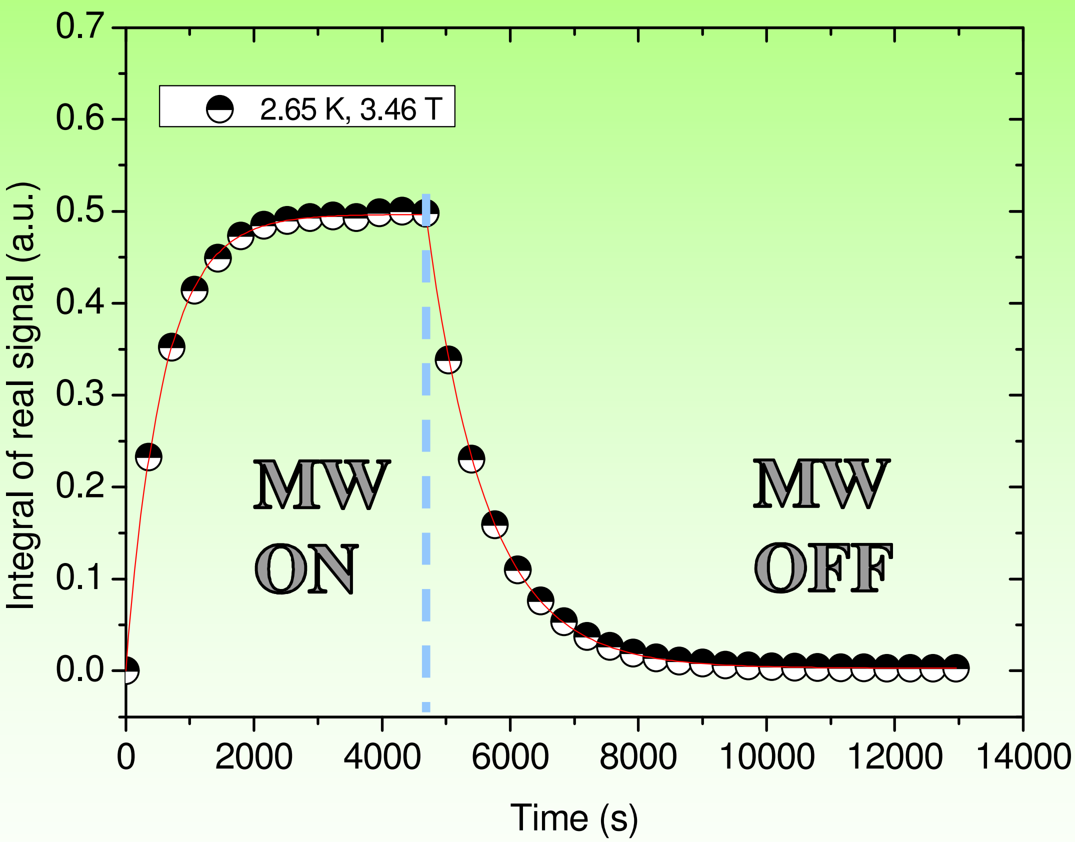
SE: non interacting electrons
TM:interacting electrons



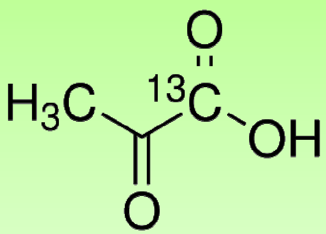
Carver, T. R.; Slichter, C. P. (1953). "Polarization of Nuclear Spins in Metals". *Physical Review* **92** (1): 212–213.

Polarization of Pyruvic acid at 3.46 T

MW source (15mW) about **96.92 GHz**
Pyruvic acid +trItyl radical 15mM



^{13}C Pyruvic Acid



- Endogenous molecule, important for the study of tumoral activity.
- NMR signal increased by a factor 100000

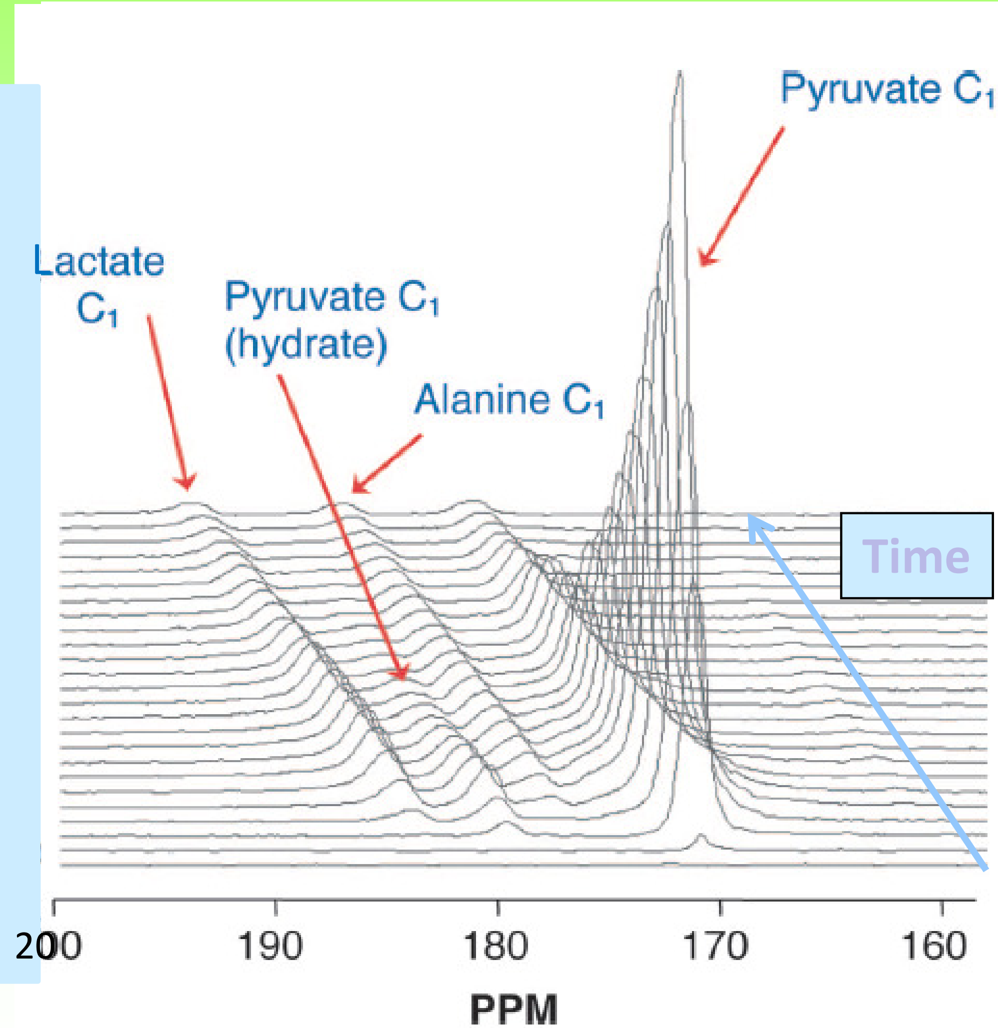
DNP for metabolic imaging

- Solutions of ^{13}C labelled metabolites and paramagnetic radicals (about 10 mM)
- Thermal mixing (TM) at low temperature (1.2 K) and high magnetic fields (3.35T)

Polarization and dissolution

Injection and MRI

Transfer line for the sample



K. Golman et al. PNAS **103** (2006), 30, 11270-11275

Magneto - therapy

<u>*Start-up clarifying requests*</u>

- Biological effects of ELF (*Extremely Low Frequency*) magnetic fields
- Mechanisms of interaction
- Definition of experimental protocol on cells

A similar system actually used by Prof. Benazzo, Policlinico PV

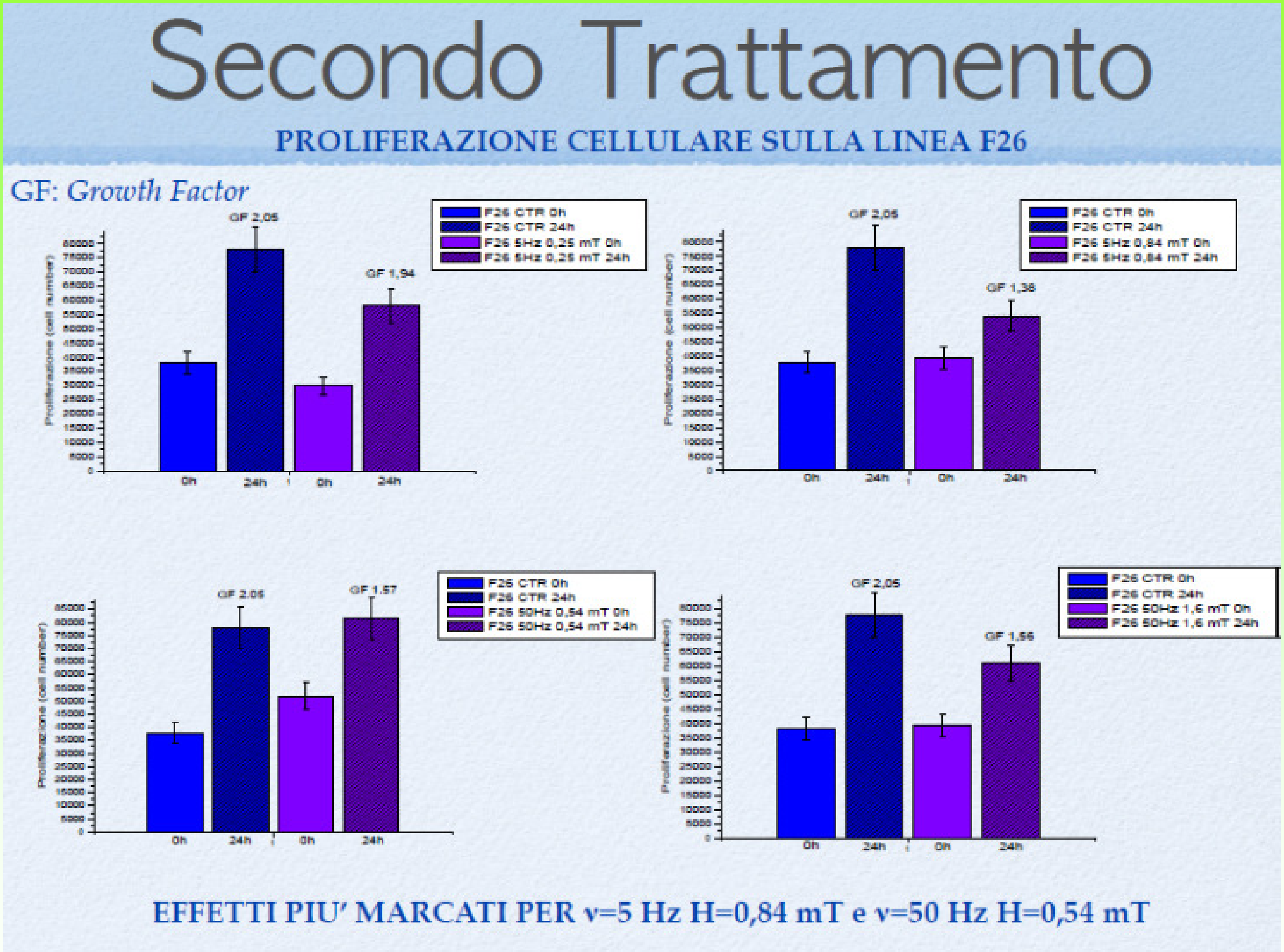
MAGNETOTERAPIA PORTATILE "FLEXA" Asalaser
Vicenza

CARATTERISTICHE TECNICHE

2 Canali con 2 uscite per applicatori
Frequenza variabile 0,5 - 50 Hz
Intensità del Campo Magnetico 0 - 1,6 mT
Durata trattamenti 0-99 minuti oppure continua
Programmi preimpostati per patologia
Possibilità di memorizzare trattamenti

APPLICATORE

Solenoide Portatile
30 cm diametro
22 cm di altezza
6,5 Kg di peso



- Effetto sulla proliferazione cellulare (fibroblasti)

The end

Any questions ??