Magnetic Nanoparticles: recent advances in biomedical applications

P. Arosio

Dipartimento di Fisica and INSTM, Università degli studi di Milano, Milano (Italy)
• Brief intro to MNPs

• State of art for the Bio-Med applications (more insight into MFH)

• Focus on NMR relaxometry (for clinical MRI):
  - Basic concepts
  - Reality vs Ideality
  - Several examples of our research

• Conclusions
What kind of magnetic nanoparticles we’re talking about?

Simplest (and mainly) form:
- magnetic core (often simple ferrites)
  + coating (variable)

Superparamagnetic NPs

What about physico-chemical properties?

\[ \text{Fe}_3\text{O}_4 \quad \gamma\text{-Fe}_2\text{O}_3 \]
Magnetic Nanoparticles

Weiss domains

Size effect ⇨ Superparamagnetism

Hysteresis curve of a ferro- or ferrimagnetic material

Giant Spin

\[ d_c = 20-100 \text{ nm} \]

Small Anisotropy Energy compared to Thermal energy

\[ L(x) = \coth x - \frac{1}{x}; x = \frac{\mu_{SPM} B_0}{k_B T} \]
Magnetic Nanoparticles

Stoner-Wolhfarth model:
The inversion of M through a coherent movement of all the spins of the particle

Energy barrier: \( E_A = k_A V \sin^2 \theta \)
- \( k_A \) = anisotropy constant, \( V \) = particle volume

Neel correlation time:
\[ \tau_N = \tau_0 \exp\left(\frac{E_A}{k_B T}\right) \]

If NPs interact:
Vogel-Fulcher model,
\[ \tau_N = \tau_0 \exp\left[\frac{E_A}{k_B (T-T_0)}\right] \]

When \( B_0 = 0 \)
Magnetic Nanoparticles

When $B_0 \neq 0$

For Biomed:
MNPs dispersed in solvents

$$1/\tau = 1/\tau_N + 1/\tau_b$$
also Brownian contribution

$\tau_b = \frac{3DH\eta}{2k_BT}$

IMPORTANT: $M_s(H)$ values, magnetic anisotropy, correlation time, field and T.
Several microscopic parameters influencing the magnetic properties of superparamagnetic NPs

- **Size** of magnetic core
- Magnetic energy and anisotropy
- Kind of magnetic ion
- **!!** Kind of coating **!!**
- Dispersant
- Shape of the nanoparticle
- Spin Topology
In particular... for **biomedical properties**

Kind of coating: **biocompatibility** and **targeting**

Surface **functionalization**

**Fluorescent/luminescent molecules**

**Drugs** “attachment” or “inclusion”

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Doxorubicine

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Taxol
The ideal task

A single **theranostic** nano-object

Diagnostics: **MRI CA**, Optical Imaging, PET, ...

Therapy: Magnetothermia (MFH), drug release
Magnetism of magnetic nanoparticles in biomedicine

Sensing

Before injection

After injection

MOVING

Magnetic hyperthermia

HEATING

Magnetic transport
**IDEA:**
IV injection + local drug release
(under external stimulus)

Magnetic transport (few preclinical examples)

**Forces on a magnetic nanoparticle:**
\[ F_m = (m \cdot \nabla) B \]
\[ F_m = \nabla \chi \Delta \chi \nabla (\frac{1}{2} B \cdot H) \]

Hydrodynamic drag force:
\[ F_d = 6 \pi \eta R_m \Delta v \]

Equating the two:
\[ \Delta v = \frac{R_m^2 \Delta \chi}{9 \mu_0 \eta} \nabla (B^2) \quad \text{or} \quad \Delta v = \frac{\xi}{\mu_0} \nabla (B^2) \]

Labelling of stem cells with MNPs

Extravasal circulation

Prototype:
250 ml water with 5.6 mg/ml Endorem pumped at 60 ml/h for 3 h in the presence of a permanent magnet

In vitro labelling of Ntera2 stem cells with red fluorescent-conjugated Bangs particles
DAB-enhanced Prussian blue staining for iron following in vitro labelling with Endorem

... or use of magnetic field gradients for drug delivery
MFH treatment

Magnetic Fluid Hyperthermia (MFH) or Magnetothermia

**Heating** through application of **AC magnetic field** via activation of **MNPs** directly implanted in the tumour mass at high doses (ca. 50 mg/cm³)

Typically in clinics: \( \nu \sim 100 \text{ kHz} \), amplitude 10 kA/m

**Minor side effects**

See M. Avolio and M Cobianchi posters for details
**MFH: Clinical applications on Glioblastoma**

- MNPs coated with amminosilane
- Direct injection in the brain tumour
- Tumour cells
- AMF
- Heating – kill tumour cells

Started a new study on glioblastoma multiforme in 2014
Several german hospitals involved
HADROCOMBI: Combining Hadron Therapy with Magnetic Hyperthermia

(A) targeted proton therapy deposits most energy on target
(B) conventional radiation therapy deposits

Illustration of MFH concept

Investigation of the possible combined action of the two therapeutic techniques, for going one step beyond the state of art of pancreatic cancer therapy. X-rays irradiation will be used as control and comparison technique.
The Sentimag® is a Class IIa device, **CE-approved for marketing and sales in Europe**, and TGA-approved for Australasia.

**Key features and benefits of Sienna⁺®:**

- Particle size optimised for filtration and retention by sentinel lymph nodes
- Simple storage and handling procedure, and significantly improved workflow compared with radioactive tracers
- Localisation can start after only 20 minutes following injection
- Natural dark brown colour eliminates the need for separate dye injections
- Non-toxic, aqueous suspension dissipates naturally in the body
- Long shelf life
- Uniquely designed and calibrated for use with Sentimag®
- Compatible with Sysmex’s One-Step Nucleic Acid Amplification (OSNA) assay (http://www.sysmex-lifescience.com/OSNA-assay-for-lymph-nodes-175-2.html)

**Sentinel lymph nodes Technique (e.g. breast cancer surgery)**
Magnetic Particle Imaging – MPI (preclinical)

It images the distribution of MNPs in biological tissues

**MNPs are tracers** and not just supportive contrast agents

1st MPI system (Bruker-Philips, 2013)

http://www.philips.com/e/imalytics/productsnew/magneticparticle.html
The most famous application of MNPs:

**T₂-negative MRI contrast agents**

Typical MRI apparatus for clinical use -> $H = 1.5$ Tesla

Prototypical example: liver tumour

Note: MNPs also as $T₁$-positive agents (see e.g. Mn-ferrites)
... and more dual or multiple diagnostic probes. Many "lab" examples.
Most of new CA for MRI are "non-specific" (i.e. not targeting) and so, two crucial questions...

1) Fate of the MNPs? 
Mostly in liver if MNPs are not reduced in total size (and not only ... ... all the physico-chemical properties of MNPs are involved !!!)

2) Medical doctors are really interested? 
or they just point to specific (i.e. targeting) or multifunctional CA ??

ALERT : SAFETY & TOXICITY !!
The MRI image intensity (the contrast) thus depends on:

**Intrinsic Parameters**
- Local proton density $N(H)$ (water, fat)
- Nuclear Relaxation times $T_1$ and $T_2$
- Magnetic susceptibility differences

**Extrinsic Parameters**
- Magnetic field
- Timing of the pulse sequence
- **Contrast Agents (CA)**

with CA the nuclear relaxation times change
(much better idea than protons’ density)

Better image contrast and pathology evidence
Focus on MRI/NMR

Fluctuations of the MNPs dipolar local field induce our local probe relaxation:

Via HYPERFINE INTERACTION

LOCAL MAGNETIC FIELDS AND DYNAMICS can be studied with NMR experimental parameters:

spectrum, nuclear spin-spin relaxation time $T_2$ and nuclear spin-lattice relaxation time $T_1$

$$1/T_1 \propto \chi T \cdot J_e(\omega_L); \quad 1/T_2 \propto \chi T \cdot J_e(0)$$

and the EFFICIENCY of a CA is:

$$\frac{1}{T_{i,oss}} = R_{i,oss} = \frac{1}{T_{i,d}} + r_i c$$

nuclear relaxivity $r_i$ ($i=1,2$) represents the increase of nuclear relaxation rate of hydrogen nuclei in presence of 1mM of magnetic center
**Focus on NMR**

**Mostly used models** for nuclear relaxation in function of size (diluted SP-NPs)

- Low anisotropy
  - Roch, Muller, Gillis, 1999
  - Levy et al, 2013
  - Fast magnetic fluctuations

- High anisotropy
  - Roch et al (clusters)
  - Slow magnetic fluctuations

- 20 < d < 40/50 nm
- d < 20 nm
- d > 50 nm

Normally we consider **core d<20 nm & spherical** shapes
(a **compromise**: good MFH efficiency and feasible targeting)

**ALERT**

- **Models tested for the** longitudinal nuclear relaxation rate $1/T_1$
- **We performed first** (to our kn.) experimental complete tests for the transverse nuclear relaxation rate $1/T_2$ (Milano and Mons group)
- **Simplified model for** $1/T_2$ (Vuong, Gossuin, Sandre et al) -> $T_2$ is the crucial parameter !!!!!!!!!!!!!!!
Typical Relaxometry curves

Analytical exact model:

$$1/T_{1,2} = f(\gamma_e, \gamma_n, \omega_L, \omega_L^n, \tau_{S1,2}, \tau_R, \tau_M, q, r, \tau_{S0}, \ldots)$$

only for small number of spins

Heuristic "approximate" expressions for nuclear relaxation rates

$$1/T_1 = (32\pi/135\,000)\mu_{sp}^2 \gamma_t^2(N_a C/RD)$$

$$\times \{7PL(x) / xJ^F[\Omega(\omega_S, \omega_0), \tau_D, \tau_N] + 7QL(x) / xJ^F(\omega_I, \tau_D, \tau_N) + 6QL(x) / xJ^F(0, \tau_D, \tau_N) + 1L^2(x) / x[3J^F(\omega_I, \tau_D, \tau_N) + 4J^F(0, \tau_D, \tau_N)] - 2L(x) / x\}$$

$$1/T_2 = (16\pi/135\,000)\mu_{sp}^2 \gamma_t^2(N_a C/RD) \{13PL(x) / xJ^F[\Omega(\omega_S, \omega_0), \tau_D, \tau_N] + 7QL(x) / xJ^F(\omega_I, \tau_D, \tau_N) + 6QL(x) / xJ^F(0, \tau_D, \tau_N) + 1L^2(x) / x[3J^F(\omega_I, \tau_D, \tau_N) + 4J^F(0, \tau_D, \tau_N)] - 2L(x) / x[3J^F(\sqrt{2\omega_I \tau_D}) + 4J^F(0)]\}$$

Crucial: dist. min approach, magn. anisotropy, $$\tau_N$$, $$M_s$$, $$\tau_D$$, Langevin, .....
Changing the magnetic core $d$: $r_1$ heuristic fit model works

NMR-D study of the local spin dynamics and magnetic anisotropy in different nearly monodispersed ferrite nanoparticles

Free parameters: $r$ (minimum approach distance), $\tau_N$, $P&Q$ (weight of magnetic anisotropy)

COLLABORATIONS
Dept. of Chemistry and INSTM, Univ. of Firenze (Italy): C. Sangregorio (CNR), C. Innocenti, E. Fantechi, A. Caneschi, D. Gatteschi
Dept. of Chemistry and INSTM, Univ. of Cagliari (Italy): M. F. Casula, P. Floris
Montpellier University (France): Y. Guari, J. Larionova
Nantes University (France): E. Ishow, L. Lartigue
* Regarding **magnetization reversal**: 

“local” $\tau_0$ and anisotropy barrier $E_A$, i.e. info about 

“local” Neel correlation time $\tau_N$ 

(comparison with $\chi_{AC} \rightarrow$ bulk) 

* The **distance of minimum approach**. This is 

influenced by coating/functionality of the sample, and **often “ignored”** in models. 

Comparison with AFM, DLS and TEM data.

* Information on **magnetic anisotropy**.
Changing the magnetic core: $r_1$ ok, but $r_2$ ...

First complete experimental $r_2$-relaxivity profile

**Theoretical NMR-D curves**

SAMPLE

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<table>
<thead>
<tr>
<th>Sample</th>
<th>Composition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Co8Ni0</td>
<td>Co$<em>{0.82}$Fe$</em>{0.18}$O$_4$</td>
</tr>
<tr>
<td>Co6Ni3</td>
<td>Co$<em>{0.63}$Ni$</em>{0.37}$Fe$_{0.29}$O$_4$</td>
</tr>
<tr>
<td>Co4Ni5</td>
<td>Co$<em>{0.42}$Ni$</em>{0.58}$Fe$_{0.29}$O$_4$</td>
</tr>
<tr>
<td>Co2Ni8</td>
<td>Co$<em>{0.17}$Ni$</em>{0.83}$Fe$_{1.09}$O$_4$</td>
</tr>
<tr>
<td>Co0Ni10</td>
<td>Ni$<em>{0.09}$Fe$</em>{1.90}$O$_4$</td>
</tr>
</tbody>
</table>

SAME FIT

Dashed = $r_2$

PARAMETERS Solid = $r_1$

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**COLLABORATIONS**

Dept. of Chemistry and INSTM, Univ. of Firenze (Italy): C. Sangregorio (CNR), C. Innocenti, E. Fantechi, A. Caneschi, D. Gatteschi

Dept. of Physics and INSTM, Univ. of Pavia (Italy): M. Corti
... maghemite, model for $r_2$ does not work again

Local spin dynamics of iron oxide magnetic nanoparticles dispersed in different solvents with variable size and shape: A $^1$H NMR study

M. Basari, T. Orlando, P. Aroldo, M. F. Casula, D. Espo, S. Murgia, C. Sangregorio, C. Innocenti, and A. Lascialfari

COLLABORATIONS
Dept. of Chemistry and INSTM, Univ. of Firenze (Italy): C. Sangregorio (CNR), C. Innocenti, E. Fantechi, A. Caneschi, D. Gatteschi
Dept. of Chemistry and INSTM, Univ. of Cagliari, Cagliari (Italy): M. F. Casula, P. Floris
A really simplified model for $T_2$

Simplifying the heuristic expression:

\[
\frac{r_2}{[Fe]} = \frac{4\gamma^2 \mu_0^2 \nu_{mat} M_0^2 d^2}{405 D}
\]

which is valid only if the Redfield condition is fulfilled:

\[\Delta \omega_0 < 1 \quad \text{MAR}\]

\[
\frac{r_2^*}{[Fe]} = \frac{2\pi \gamma \mu_0 \nu_{mat} M_0}{9\sqrt{3}} \approx r_2
\]

SDR

But for a complete theory a more refined model is needed!

Study in progress…… influence of interparticle interactions, microaggregation, water (exchange-penetration & coating interaction with bulk) role, Brownian motion (if…),…….
**Other cases**

**Functionalization effect**

Superparamagnetic iron oxide nanoparticles functionalized by peptide nucleic acids

Marco Galli, Andrea Guerrini, Silvia Cauteruccio, Pramod Thakare, Davide Dova, Francesco Orsini, Paolo Anseli, Claudio Carrara, Claudio Sangregorio, Alessandro Lascaletti, Daniela Maggiom and Emanuela Licandro

**Hollow topology**

Surface vs bulk Spins

\[ \frac{1}{T_1} = A'HF \left( \frac{g_0}{1 + \omega^2 \tau_0^2} \right) + (1 - A'\gamma) \frac{1}{T_1^{\text{Heu}}} \]

\[ \frac{1}{\tau_c} = \frac{1}{\tau_{SLPM}} + \frac{1}{\tau_D} \]

 Changes of \( r_i \)

**paramagnetic contribution**

(also other evidences: magnetic, MuSR, ...)

See M. Basini poster

**COLLABORATIONS**

Dept. of Chemistry and INSTM, Univ. of Milano: G. D’Alfonso, D. Maggioni, E. Licandro, et al

**COLLABORATIONS**

ISM-CNR, Roma: D. Peddis, et al
MR imaging and targeting of human breast cancer cells with folate decorated nanoparticles

MR images about targeting: towards molecular imaging

* MDA-MB-231 human breast cancer
* Subcutaneous implantation

NPs with folic acid

NPs without folic acid

Endorem
Conclusions (not exhaustive)

* Nowadays For ferrites $d > 10 \text{ nm}$ is ok (well joint to $d > 14 \text{ nm}$ for MFH), but the size is crucial for bio-application -> reducible?

* Surface spins/Solvent/Coating effects to be clarified

* Role of interparticle interactions? Theoretically manageable?

* Need for specific model if functionalization with drugs, fluo molecules, antibodies/peptides, are implemented

* Industrial scalability (stimulate companies interest)

* Control Protein Corona effect and avoid (except specific cases) macrophages actions

* Poor specific uptake in tumor tissue proved. Percentage enough for ….?

* Cells mechanism of uptake and EPR (Enhanced Permeability and Retention) effect

* Problems of haemagglutination and aggregation

* Toxicity has to be established case by case

Crucial passage for new systems is from in-vitro to in-vivo !!!
Our Group

The boss

The old pillars

The workers
Comparison of different kinds of Hyperthermia

Heating by microwave and radiofrequency sources
- good localization at shallow depths
- Weaknesses: (i) cannot become selective; (ii) high temperature also all around in normal tissues; (iii) at greater tumor depths, even with lowered frequency, the localization is much poorer; (iv) invasivity of the implant; (v) many repeated treatments (thermo-tolerance)

Heating by ultrasound sources (and HiFUS)
- good penetration and temperature can be achieved in soft tissues
- Weaknesses: (i) cannot become selective; (ii) high temperature also all around in normal tissues (MRI confirms); (iii) the presence of bone or air cavities causes distortions of the heating pattern; (iv) many repeated treatments (thermo-tolerance)

Magnetic Fluid Hyperthermia
- Advantages of MFH: (i) Innovation: joint to Hadron-therapy first time (in literature, in combination just with radiotherapy); (ii) local temperature increase/control, normal tissues negligibly affected; (iii) no implant invasivity; (iv) less theoretical limitation vs kind of tumour; (v) single injection also for repeated treatments; (vi) tumor reachable at greater depth; (vii) in perspective the magnetic nanoparticles can carry a drug or antibodies, peptides, etc. (MFH can become selective)
- Weaknesses: high MNPs doses (from literature no short/medium-term major side effects), inhhomegeneity of MNPs spatial distribution
**Specific Absorption Rate (SAR)**

SAR: the rate at which energy is adsorbed by the body when exposed to a radio frequency (RF) electromagnetic field (generally 100 kHz÷1 GHz).

It is also called SLP (Specific Loss Power)

It is defined as the power absorbed per mass of tissue (W/kg).

SAR can be calculated from the electric field or the magnetic field within the tissue as:

\[
\text{SAR} = \int_{\text{sample}} \frac{\sigma(r)|E(r)|^2}{\rho(r)} \, dr
\]

\[
P_{FM} = \mu_0 f \int H \, dM
\]

(SAR \(\equiv P_{FM}\))

where \(\sigma\) is the sample electrical conductivity, \(E\) is the RMS electric field, \(H\) the RMS magnetic field, \(f\) the frequency of \(H\), \(M\) the magnetization, \(\rho\) is the sample density.

In **magnetic hyperthermia** is expressed in W/g of nanoparticles:

(i) \(\text{SAR} = A \cdot f\) (hysteresis losses) or (ii)

(ii) \(\text{SAR} \propto f \chi''(t) H_0^2\) (relaxation losses, Brownian/Neel)

where \(A = \text{area of the hysteresis loop}\) and \(f = \text{frequency of the rf magnetic field}\).

In the case of MNP, \(A\) depends on \(K, V, T, f, H_0, c\).
Coming back to the origin

**Typical dimensions in biomedicine**

- Cells
- Lymphocytes
- Erythrocytes
- DNA
- Proteins
- Lipid bilayer
- Quantum dots
- Bucky balls
- Small molecules and atoms
- Dendrimers
- Micelle
- Drug
- Emulsion
- Nanoparticulate systems
- Liposomes

**Scale**
- $10^{-3}$ m (millimeter)
- $10^{-6}$ m (micrometer)
- $10^{-9}$ m (nanometers)
- $10^{-12}$ m (picometer)

**Spleen cut-off**
- 200 nm

**Nanoparticles used in drug delivery**
<table>
<thead>
<tr>
<th>Why MRI?</th>
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<tbody>
<tr>
<td><strong>Nuclear Medicine:</strong></td>
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<tr>
<td>• Poor spatial resolution</td>
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<tr>
<td>• Poor temporal resolution</td>
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<tr>
<td>• High sensitivity</td>
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<tr>
<td>• Reporters: radionuclides</td>
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<tr>
<td><strong>Optical Imaging:</strong></td>
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<tr>
<td>• Reporters: luminescent probes</td>
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<tr>
<td><strong>X-Ray (CT):</strong></td>
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</tr>
<tr>
<td>• Low sensitivity</td>
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</tbody>
</table>

**MRI:**
- **Non-invasive**
- Good spatial resolution
- Good temporal resolution
- Low sensitivity
For biomed: our roles let us to

- Help the chemists
  Accurate study of Chemico-Physical properties of MNPs -> choice of better synthetic pathways to follow

- Understand the hopeful application

Diagnostics
  CAs

Therapeutics
  Hyperthermia/ drug delivery

... again
If “tumour (disease) targeting” at the level of clinical applications is actually almost **prohibitive**, what could be the “industry” and clinicians interests?

Still obtaining a “**small**” non-specific CA, with well controllable synthesis and with **efficiency** (**relaxivity**) **higher** than actual ones (**lower costs, lower doses**) BUT SAFE!

This “guides” the research about controlling the **physical mechanisms/parameters** that enhances the nuclear relaxation
Examples of other models for SP MNPs

Figure 1: Experimental NMRD profiles and theoretical fits (solid lines) of suspensions of (a) \(\gamma\text{Fe}_2\text{O}_3\) and (b) CoFe\(_2\text{O}_4\) NPs (iron concentration \([\text{Fe}] = 1 \text{ mM}\)). The fits are obtained as explained in the text using \(T = 298 \text{ K}, \eta = 0.89 \times 10^{-3} \text{ Pa s}\) and \(M_s\) the bulk value (412 kA m\(^{-1}\) for \(\gamma\text{Fe}_2\text{O}_3\) and 398 kA m\(^{-1}\) for CoFe\(_2\text{O}_4\)). The adjustable parameters used for each sample are the following. For FeO1: \(K = 9000 \text{ J m}^{-3}, \tau_0 = 6 \times 10^{-8} \text{ s}, \alpha = 0.11\) and \(\delta = 1.8\) nm. For FeO2: \(K = 6000 \text{ J m}^{-3}, \tau_0 = 5 \times 10^{-9} \text{ s}, \alpha = 0.19\) and \(\delta = 1.4\) nm. For CoFe1: \(\delta = 1.5\) nm. For CoFe2: \(\delta = 0.5\) nm. Insets show the normalized magnetization curves fitted with polydisperse Langevin functions (solid lines) to determine the characteristic diameter \(d_0\) and polydispersity \(\sigma_d\) of the NP log-normal size distributions. We found: for FeO1: \(d_0 = 9.2\) nm, \(\sigma_0 = 0.22\); for FeO2: \(d_0 = 7\) nm, \(\sigma_0 = 0.2\); for CoFe1: \(d_0 = 7.9\) nm, \(\sigma_0 = 0.32\) and for CoFe2: \(d_0 = 6.2\) nm, \(\sigma_0 = 0.4\).
Ferumoxytol in Clinical Practice: Implications for MRI

Brendan J. McCullough, MD, PhD, Orpheus Kolokythas, MD, Jeffrey H. Maki, MD, PhD, and Douglas E. Green, MD

Figure 1. T1 shortening from ferumoxytol results in blood pool hyperintensity, obscuring enhancement from GBCA. Axial T1-weighted image prior to GBCA administration (a) and postcontrast images during arterial (b), portal venous (c), and equilibrium phases (d) show unchanged enhancement. Note is made of small esophageal varices (arrows/asterisks).

Figure 2. Renal cortical enhancement confirms appropriate administration of GBCA. Axial T1-weighted image demonstrates homogeneous signal intensity of the kidneys before administration of GBCA (a). Following injection of GBCA (arterial phase), there is perceptible enhancement of the renal cortex (b).

Figure 3. Axial T2-weighted single-shot image through the liver and spleen demonstrates hypointensity in the spleen due to the T2 shortening effect from iron accumulation. Ferumoxytol is taken up by the reticuloendothelial system, presumably resulting in the observed iron accumulation.

Figure 4. Axial out-of-phase image (TR 2.5 ms, TE 4.6 ms) shows T1 hyperintensity in the blood pool (a). Axial in-phase image (TR 4.6 ms) shows pronounced signal dropout in the spleen due to the T2* effect from iron accumulation, presumably from ferumoxytol uptake (b). No significant signal loss is observed in the liver on the in-phase image, indicating little or no ferumoxytol uptake.
one of examples of non-specific CA

MRI with Co-ferrites (Colorobbia)
liver of normal rats, at 1 day from the bolus injection

Even just our group collaborated with several researchers synthesizing novel MNPs with high transverse relaxivity (i.e. efficiency in MRI image contrast) until 8 times the (ex-)commercial compound Endorem
Other images about targeting and ... shape

Superparamagnetic iron oxide nanoparticles for in vivo molecular and cellular imaging
Shahriar Sharifi\textsuperscript{a,b}, Hajar Seyyednejad\textsuperscript{b}, Sophie Laurente\textsuperscript{c,\textsuperscript{d}}, Fatemeh Atyabi\textsuperscript{a}, Amir Ataei\textsuperscript{a,f} and Morteza Mahmoudi\textsuperscript{b,\textsuperscript{d,\textsuperscript{e}}}.

* Single Chain Antibody Fragments

Water-Dispersible Ferrimagnetic Iron Oxide Nanocubes with Extremely High $r_2$ Relaxivity for Highly Sensitive in Vivo MRI of Tumors
Nohyun Lee,\textsuperscript{e,\textsuperscript{f}} Yoonseok Choi,\textsuperscript{e,\textsuperscript{f}} Youjin Lee,\textsuperscript{b} Mihyun Park,\textsuperscript{b} Woo Kyung Moon,\textsuperscript{c} Seung Hong Choi,\textsuperscript{e,\textsuperscript{f}} and Taeghwan Hyeon\textsuperscript{e,\textsuperscript{f}}.

* Nude mouse with melanoma
* Cube

Figure 4. In vivo MR Images of the tumor site before (a) and 1 h after (b) intravenous injection of WFIONs (arrows indicate the tumor sites). After administration of WFIONs, the MR signal of the tumor is significantly attenuated.
Antibody-Functionalized Magnetic Polymersomes: In vivo Targeting and Imaging of Bone Metastases using High Resolution MRI

Line Pourtau, Hugo Oliveira, Julie Thevenot, Yali Wan, Alain R. Brisson, Olivier Sandre, Sylvain Miraux, Eric Thiaudiere,* and Sébastien Lecommandoux†

Figure 3. Bone BT-474 tumor targeting as assessed from high resolution 3D TrueFisp MRI. Extracted axial views and color map of relative signal change in percent brought by the injection of naked (A) or targeted (B) polymersomes. Longitudinal views before (C, D) and after (E, F) injection of naked (C, E) and targeted (D, F) polymersomes. Red arrows denote tumor tissue. White arrows denote contrast variations on tumor boundaries. Experiments were performed when the tumors reached a volume of 12 to 15 μl.

within EU- FP7-Nanother
TARGETING: a different approach

Bacteriophage as a scaffold for MNPs

Modified M13 filamentous bacteriophage with MNPs ($r_2 = 35 \text{ mM}^{-1}\text{s}^{-1}$) and a targeting peptide

Target: SPARC glycoprotein

C4-2B prostate cancer cell line

Ghosh et al.

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**Figure 3 | Targeting in vivo using MRI and correlative histology.** a,b, MR scans of mice with C4-2B tumours (encircled) pre-injection and 24 h post-injection, respectively, with M13-SBP-MNP. c,d, MR scans of DU145 control tumours (circled) pre-injection and 24 h post-injection with probe, respectively. Note the maintenance of the bright image of the tumour (circled) in DU145 pre- to post-injection, whereas a post-injection dark contrast against the pre-injection bright MR image is observed in C4-2B (circled). All tumours formed subcutaneously in athymic nude mice and were imaged using a 7 T small animal MR.
3 main parameters:

- spectrum
- nuclear spin-spin relaxation time $T_2$
- nuclear spin-lattice relaxation time $T_1$

Local Probe

- Nuclei are local probes $\Leftrightarrow$ sensitive to local hyperfine interactions
- Local spin dynamics (mainly $T_1$ and $T_2$) and spin distribution (mainly spectra) can be studied

In MRI and relaxometry, sensitivity to spin dynamics and molecular “motion”
Nuclear Relaxation Mechanisms

\[ R_{1,2,\text{oss}} = \frac{1}{T_{1,2,d}} + \frac{1}{T_{1,2,\text{inner}}} + \frac{1}{T_{1,2,\text{outer}}} \]

\[ 1/T_{1,2} = f(\gamma_e, \gamma_n, \omega_L^e, \omega_L^n, \tau_{s1,2}, \tau_R, \tau_M, q, r, \tau_{s0}, \ldots) \]

**Several correlation times** within the game:

- Chemical exchange time of coordinated water \( \tau_M \)
- Rotational time (brownian) \( \tau_R \)
- Electronic relaxation time (also Neel reversal) \( \tau_{Si} \)
- Diffusion time \( \tau_D \)

Two main contributions

Inner Sphere (IS)

Outer Sphere (OS)
Colloidal assemblies of oriented maghemite nanocrystals and their NMR relaxometric properties

Athanassia Kostopoulou, Sabareesh K. P. Velu, Kalaivani Thangavel, Francesco Orsini, Konstantinos Brintakis, Stylianos Psycharakis, Anthi Ranella, Lorenzo Bordonali, Alexandros Lappas and Alessandro Lascialfari

Assemblies of oriented maghemite nanocrystals

$r_2 > 400-500 \text{ mM}^{-1}\text{s}^{-1}$
Protein corona affects the relaxivity and MRI contrast efficiency of magnetic nanoparticles

Houshang Amiri, Lorenzo Bordonali, Alessandro Lascialfari, Sha Wan, Marco P. Monopoli, Isseult Lynch, Sophie Laurenç and Morteza Mahmoudi

\[ r_2 > 100-200 \text{ mM}^{-1}\text{s}^{-1} \]

Protein corona affects \( r_2 \) !!

* Plain \( \Rightarrow \) no

* "–“ charge \( \Rightarrow \) slight increase

* " + “< charge \( \Rightarrow \) decrease
Magnetic Fluid Hyperthermia (MFH)

...... after and/or trying to go beyond Jordan’s clinical studies
MFH: Iron/M oxide nanoparticles

Optimization of $K$, $M$ and $D$ in core-shell NPs

High SLP
Results:
- **Increase** in median OS-2 -> 7.2 months
- **Increase** in median OS-1 -> 8.6 months
- Few side effects

**major Drawbacks observed:**
- no MRI after treatment
- no metallic materials < 40cm treated area
MFH: Resovist® (commercial product)

- SPION CA for MRI
- Diameter magnetic core: 9 nm
- Diameter nanoparticle: 62 nm (core + carboxydextran)
- 62.1 kHz, 2.2 kW
- Tumour CT-26 (murine colon)

Tendency to diminution of tumour volume
MFH: a different type of MNPs

6 different kind of nanoparticles including **magnetosomes**

- Tumour cells MDA-MB-231 (breast)
- 40 mT
- 183 kHz
- 20 minutes
- From **AMB-1 magnetotactic bacteria**
- 3 treatments (alternate days)
- SAR Ch-Std: 390 W/g

In 1 case the tumour disappear

---

Alphandery et al.

| ![Graphs](https://example.com/graphs.png) | ![Graphs](https://example.com/graphs.png) |

---

**!!! Chains magnetosomes !!!**
MFH: core-shell nanoparticles

- Core of Fe and coating of Fe$_3$O$_4$
- $12 \pm 3$ nm
- 5 kA/m, 366 kHz
- SAR = 64 W/g
- Melanoma cells B16-F10
- $\Delta T = 11^\circ C$

The tumour volume increase rate slows down
An example of collaboration

TD COST Action TD1402

Management Committee

NC Chair: TBA
NC Vice Chair: TBA

Data registration in a COST project is subject to online registration and nomination acceptance by nominees.

COST Participants

TD COST Action TD1402

Trans-Domain COST Action TD1402

- Description
- Parties
- Management Committee

General Information

Science officer of the Action: Dr. Marie HOF-LEUSCHER CANVAC
Administrative officer of the Action: Ms. Marijke VAN DER SIJNCKT

Downloads

- Action Fact Sheet
- Memorandum of Understanding

Websites

- Website: http://www.cost.eu

* Content provided by a COST. Data is synchronised once per night.

Multifunctional Nanoparticles for Magnetic Hyperthermia and Indirect Radiation Therapy (RADIOMAG)

The Action aims to bring together and to organise the research outcomes from the different participating network members in a practical way to provide clinicians with the necessary input to trial a novel anti-cancer treatment combining magnetic hyperthermia and radiotherapy, also identifying future research objectives upon appraisal of the obtained results. Feedback between the different working groups is essential, and it is expected that the lifetime of this Action proposal will eventually result in a compendium of best practices for magnetic hyperthermia.

RADIOMAG will generate new and strengthen the existing synergies between technical advances (thermal imaging / MRI), new treatment concepts (combined targeting radio-sensitisation and magnetic thermotherapy) and biocompatible coating in order to achieve a breakthrough in the clinical application of magnetic hyperthermia. Due to the complexity of this aim, synergies can only be achieved on a longer time frame, by means of workshops, STSMs, joint publications, common Horizon 2020 research proposals and exchange with other COST Actions (e.g. TD1004, TD1205).
Sample 15_Block-M (115/15) – average diameter $d = 130 \pm 30$ nm  
Core: magnetite. Block-M copolymer coating. Functions: drug &/or folic acid  
* All samples with Paclitaxel (PTX)  
* Two classes: with and without folic acid (folic acid is the targeting agent)

<table>
<thead>
<tr>
<th>Formulation Description</th>
<th>Composition (w/v):</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>About 8 ml (exactly 10 mg/ml) Hybrid IS19b (Argus) – Magnetite (Colorita) NPs</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Pharmaceutical form</th>
<th>Suspension</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dilution solution</td>
<td>Water</td>
</tr>
</tbody>
</table>

TUMOUR MODEL (developed BY Leitat)  
* Ten female homozygote nude mice  
* **MDA-MB-231 human breast cancer**, over-expressing folate receptors  
* Subcutaneous implantation
**Relaxometry**

**Hybrids Fe₃O₄ (also Paclitaxel)**

<table>
<thead>
<tr>
<th></th>
<th>d</th>
<th>( r_{HYD} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>BLOCK-M</td>
<td>12</td>
<td>~130</td>
</tr>
<tr>
<td>BLOCK-MP</td>
<td>12</td>
<td>~130</td>
</tr>
<tr>
<td>BLOCK-MP_FA</td>
<td>12</td>
<td>~150</td>
</tr>
</tbody>
</table>

**BLOCK-MP-FA** good \( r_2 \) relaxivity compared to commercial compound Endorem. **Promising** for applications as negative MRI contrast agent (also with Paclitaxel): **8 times higher relaxivity ! ⇒ GO ON!!!**
**in vivo MRI protocol**

**INSTM-COLORITA 15_Block-M-FA (115/15)**

Mice investigated: total 10

* 2 animals with intratumoral injection of NPs with folic acid *WITHOUT* MFH treatment
* 3 animals with intratumoral injection of NPs with folic acid *WITH* MFH treatment

The above 5 animals will be sacrificed when tumour reaches 2 cc. Liver, kidneys, spleen, tumour will be excised.

* 1 animal with slow infusion of NPs with folic acid to see targeting at 2, 24 and 48 hrs
* 1 animal with slow infusion of Endorem to see targeting at 2, 24 and 48 hrs
* 1 animal with slow infusion of NPs with folic acid to see targeting at 2, 24 hrs (to be sacrificed for histological control)
* 1 animal with slow infusion of Endorem to see targeting at 2, 24 hrs (to be sacrificed for histological control)
* 1 animal with slow infusion NPs without folic acid to see targeting at 2, 24 hrs
Biodistribution

SAGITTAL T2W IMAGES - with folic acid

Pre  Post 1h  Post 24h

NPs with folic acid

MOSTLY IN LIVER

Endorem
**Zoom on targeting**

**T2W images: post 24h on the tumour**

**NPs INSTM-Colorita with folic acid**

**NPs without folic acid**

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A semi-quantitative Analysis: $T_2$ around tumour (to be refined and quantified more properly)

* Diminishes by 15-20% in NPs with folic acid
* Diminishes by 3-4% in NPs without folic acid
NPs with folic acid vs Endorem®

AXIAL T2W IMAGES
slow infusion (400 microliters in 1h, correspondent to 250 micromol/kg)

INSTM-Colorita with folic acid (target !)

Endorem (does not target, as expected)

Pre

Post 1h

Post 24h
Physico-chemical characterization

AFM Micrographs

Block-M

Block-P

Block-MP

Block-FA

Block-M-FA

Block-MP-FA
Probes: $^1H-\mu^+$

Measure: relaxation times of Nuclear Magnetization/ Muon’s Polarization

**WE MEASURE**
The Electronic spectral density

$J_e(\omega) = \text{FT} [G(r, t)]$

• Interacting with MNP’s THEY RELAX in $t < s$

$J_e(\omega)$ IS THE PROBABILITY TO FIND AN ELECTRONIC OSCILLATION AT $\omega$

**SIMPLE CASE!!**

$J_A(\omega) = \frac{\tau}{1 + (\omega \tau)^2}$

**WE WANT**
The Electronic correlation function

$G_e(t)$ DESCRIBES EQUILIBRIUM FLUCTUATIONS OF A QUANTITY F (FIELD)

$G_{AA}(\tau) = \langle F(0)F(\tau) \rangle = \lim_{T \to \infty} \frac{1}{T} \int_0^T F(t)F(t+\tau)dt$