

Magnetic Resonance Imaging (MRI)



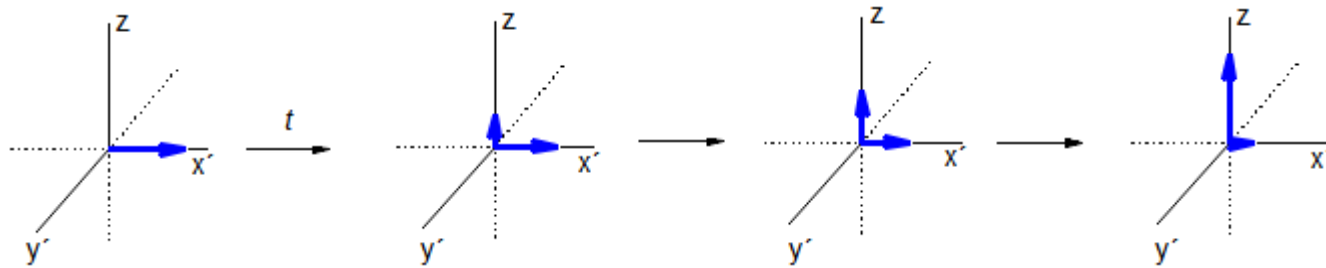
MR sagittal image of human head

- Non-invasive and safe technique
- Great spatial resolution (μm scale)
- Outstanding diagnostic capability

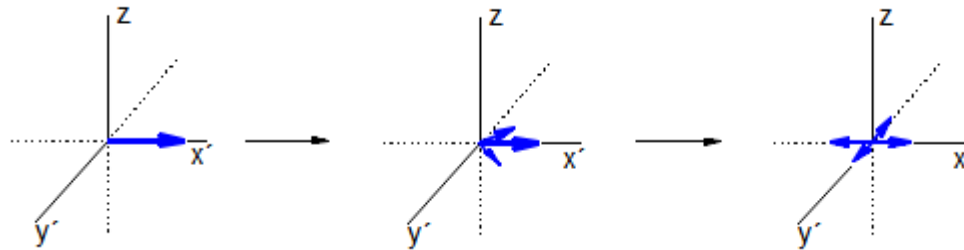
A MR-image represents a map of the intensity of the ^1H -NMR signal of water protons

The contrast is mainly generated by difference in the relaxation times (T_1 and T_2) of water protons

T_1 Relaxation (Spin-Lattice Relaxation)



T_2 Relaxation (Spin-Spin Relaxation)



$$T_1 \text{ ca. } 5 T_2$$

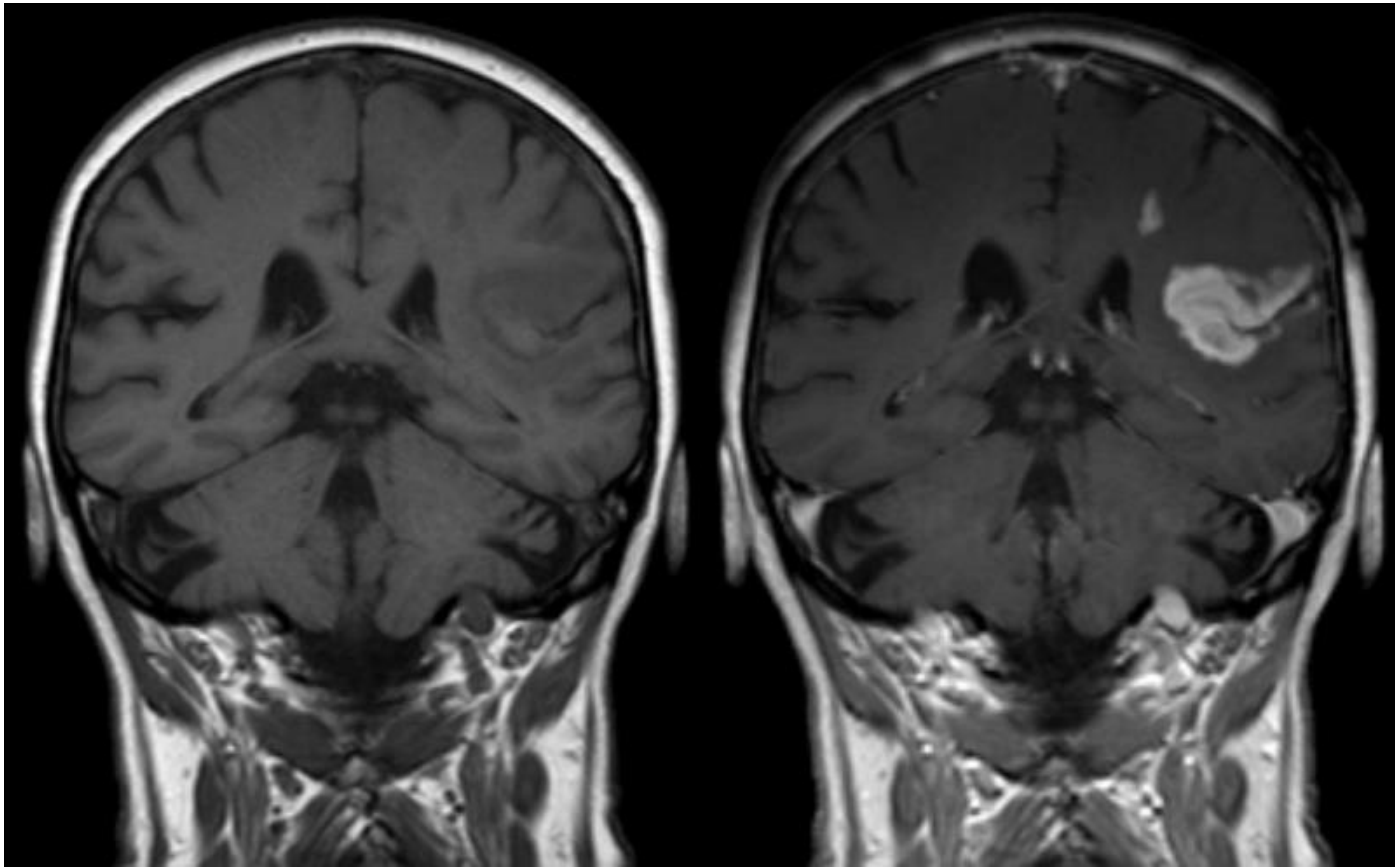
Contrast Agents (CA)

The purpose of a CA is **to reduce T_1 (parallel to B_0) or T_2 (perpendicular to B_0)** in order to obtain an hyper- or ipo-intense signal, respectively, in short times and with a better signal to noise ratio.

T_1 contrast agents (positive = hyper-intense signal):
paramagnetic metal complexes Fe(III), Mn(II),
Gd(III)

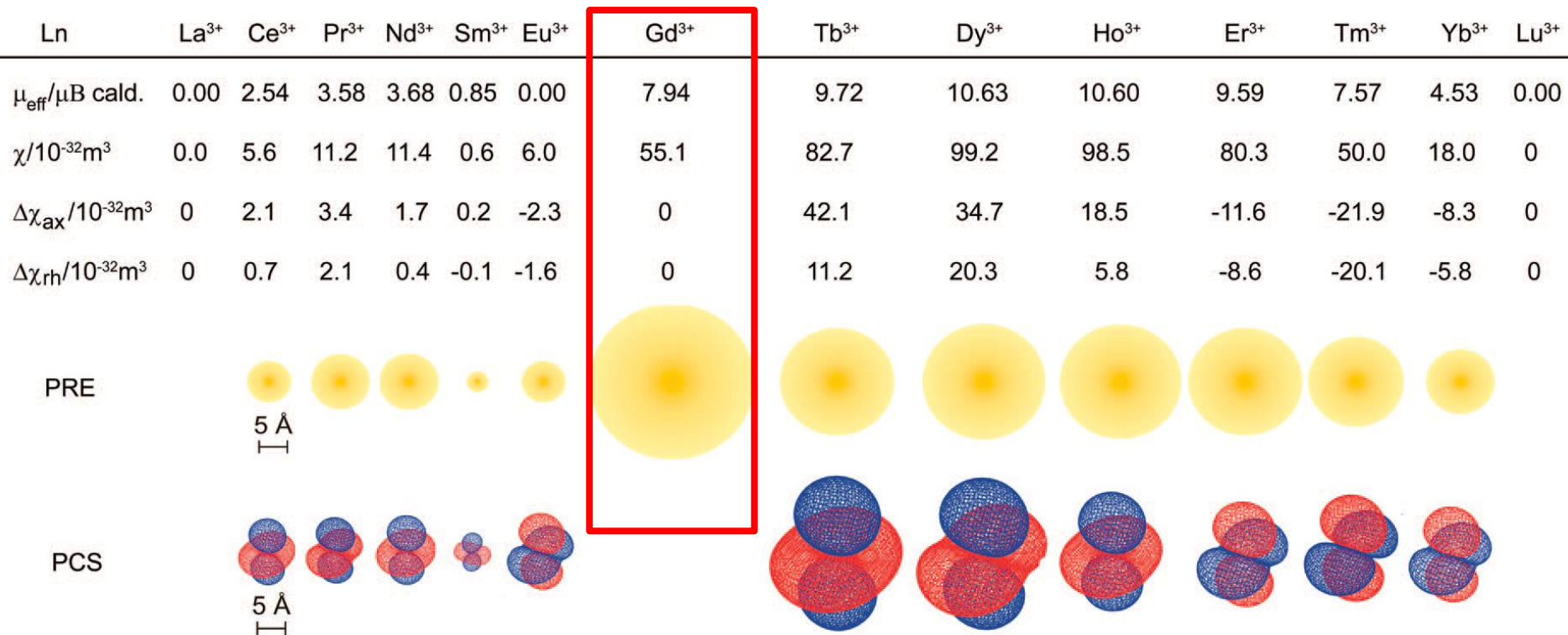
T_2 contrast agents (negative = ipo-intense signal):
Super-Paramagnetic Iron Oxide nanoparticles
(SPIO) and ultra-small super-paramagnetic iron
oxide NPs (USPIO)

MRI CA's must have a catalytic (i.e. amplified) effect
agenti extracellulari non-specifici, organo-specifici e del sangue



Defect of the blood-brain barrier after stroke shown in MRI. T1-weighted images: left image = without; right image = with contrast medium administration

Il tempo di rilassamento del momento di spin elettronico del Gd(III) è molto più lungo che per gli altri ioni lantanidici (stato di spin totalmente simmetrico)



PCS = *Pseudo-Contact Shift*

PRE = *Paramagnetic Relaxation Enhancement*

il raggio della sfera gialla indica la distanza alla quale i segnali ¹H NMR subiscono un significativo accorciamento del tempo di rilassamento

~40% MRI scans use a Gd CA

~40 million MRI scans/year use a Gd CA
worldwide

i.e. ~50 tons of Gd

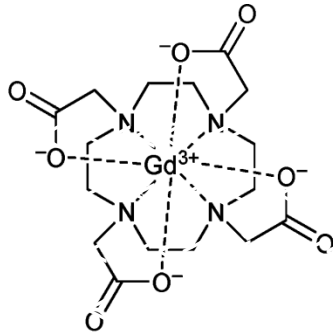
9 commercially used Gd CA

Market > 1 billion \$/year

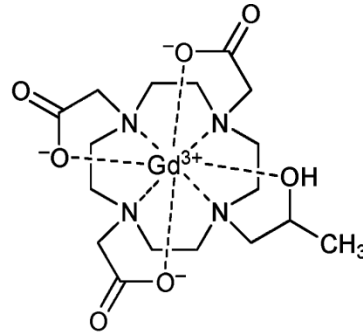
The technique has a low sensitivity: gram quantities of Gd compounds are used in each scan. This causes toxicity problems (nephrogenic systemic fibrosis)

The Gd(III) ion is quite toxic ($LD_{50} = 0.2 \text{ mmol}\cdot\text{kg}^{-1}$)

Some commercial T₁ contrast agents (extracellular fluid CAs)

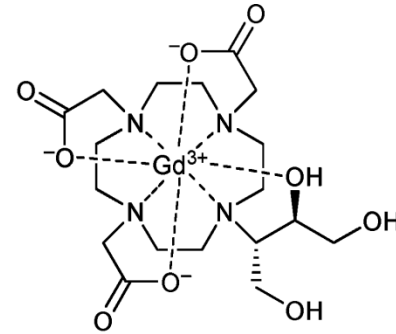


Gd-DOTA
Dotarem®
(Guerbet)

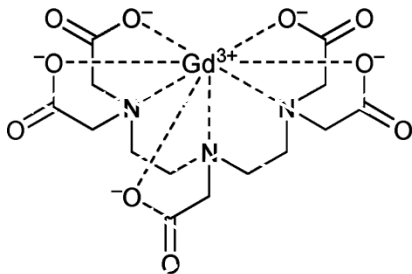


Gd-HP-DO3A
ProHance®
(Bracco)

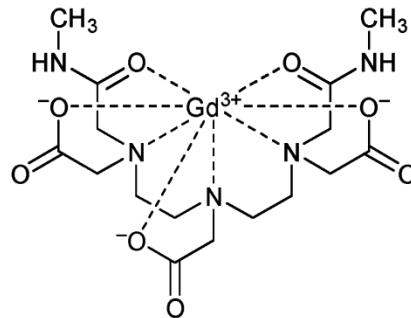
Typical dose =
0.1 – 0.3 mmoles/kg



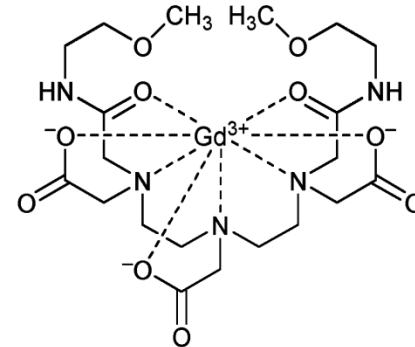
Gd-BT-DO3A
Gadovist®
(Schering)



Gd-DTPA
Magnevist®
(Schering)



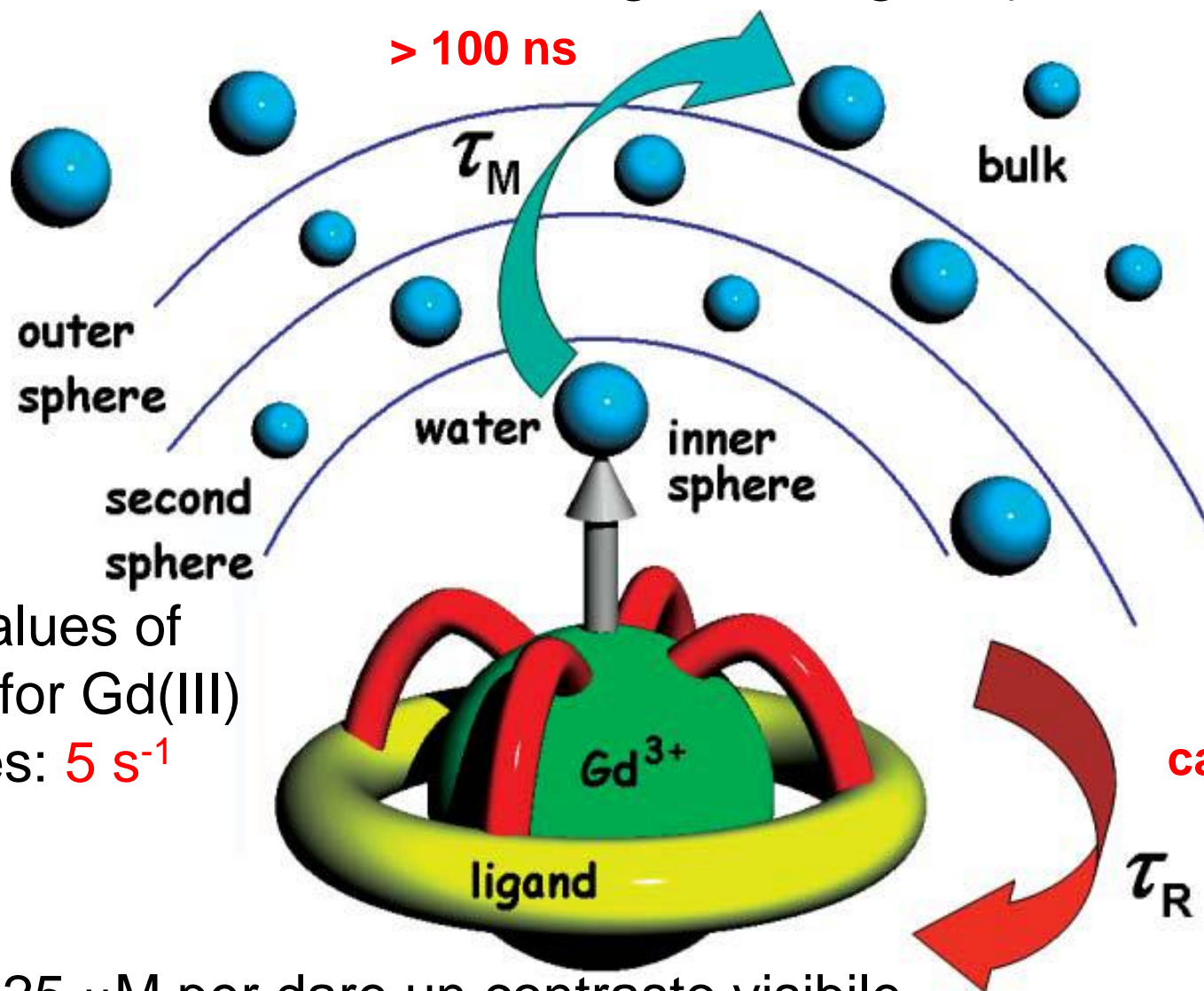
Gd-DTPA-BMA
Omniscan®
(Amersham)



Gd-DTPA-BMEA
OptiMARK®
(Mallinckrodt)

Parameters that affect Relaxivity

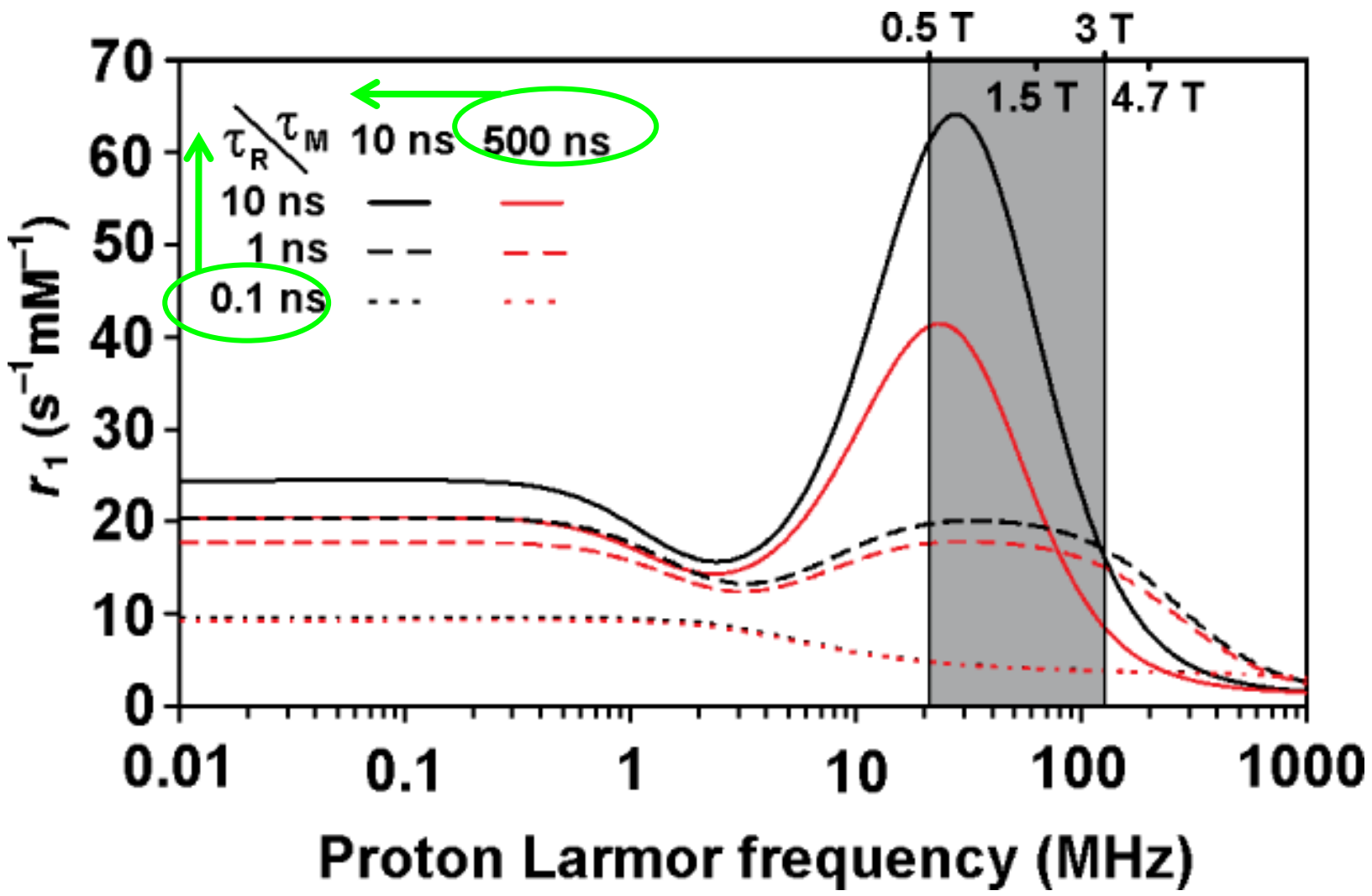
Teoria di Solomon-Bloembergen-Morgan (*SBM Theory*)



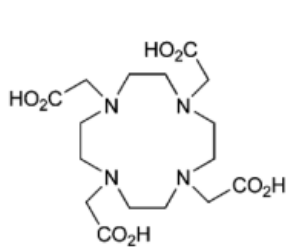
Typical values of relaxivity for $Gd(III)$ complexes: **$5 \text{ s}^{-1} \text{ mM}^{-1}$**

$[Gd] > 125 \mu\text{M}$ per dare un contrasto visibile

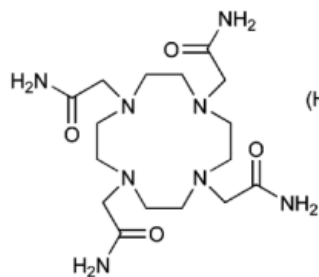
Teoria di Solomon-Bloembergen-Morgan (*SBM Theory*)



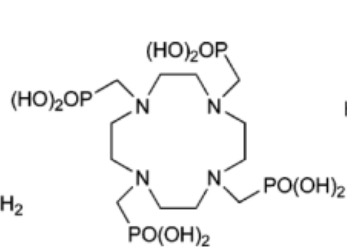
DOTA family



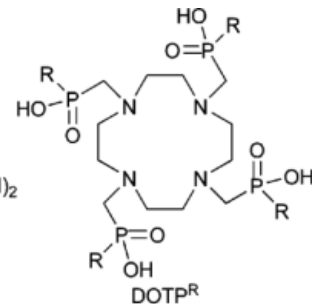
DOTA



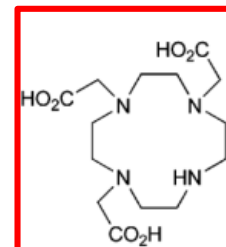
DOTAM



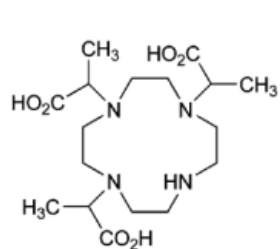
DOTP



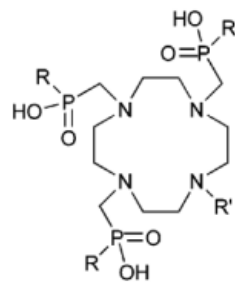
DOTP^R



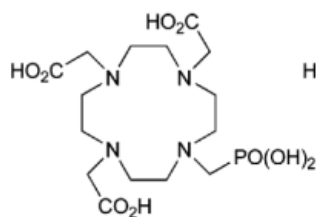
DO3A



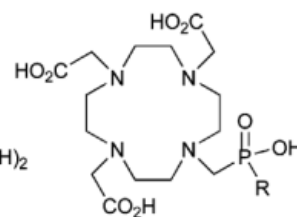
DO3MA



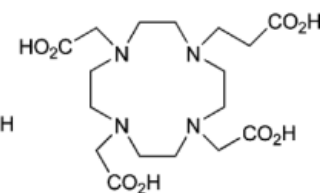
DO3P^R



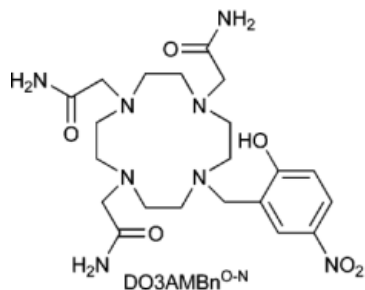
DO3AP



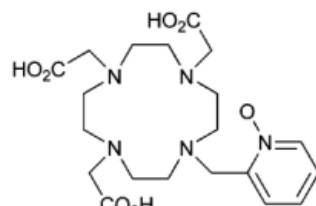
DO3AP^R



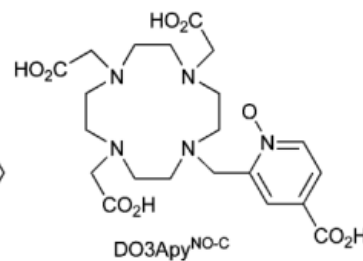
DO3ACE



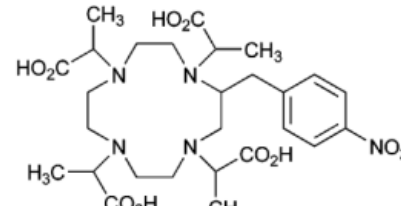
DO3AMBn^{O-N}



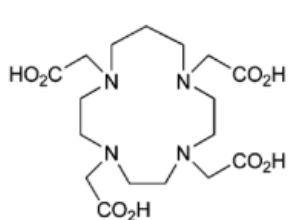
DO3Apy^{NO}



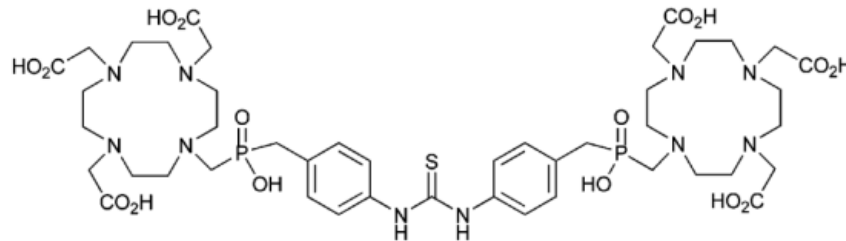
DO3Apy^{NO-C}



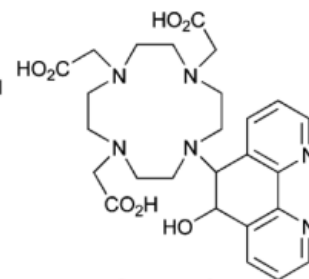
NBnDOTMA



TRITA

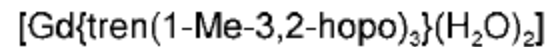
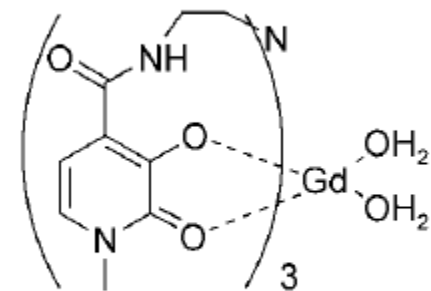
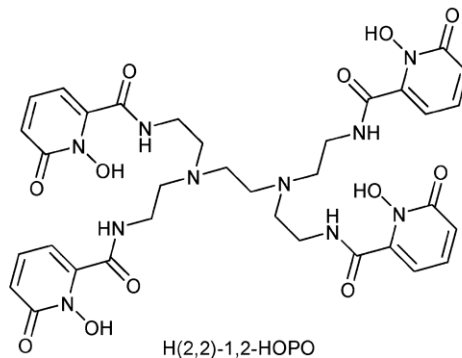
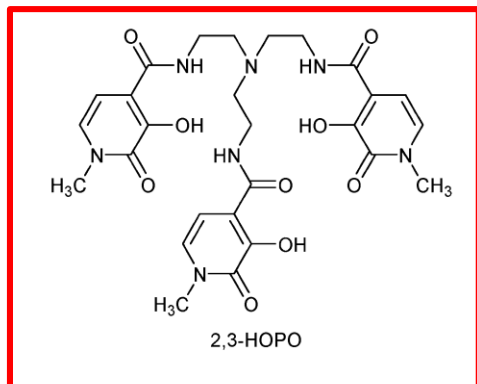
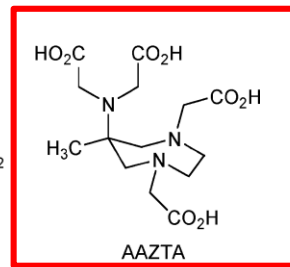
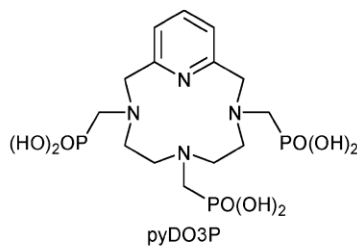
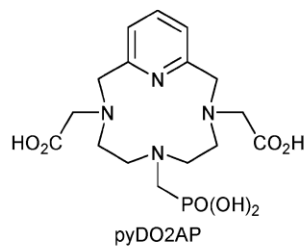
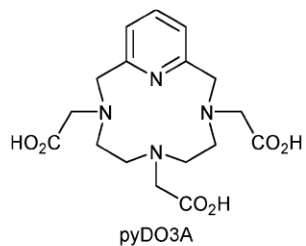


(DO3AP^{ABn})₂CS

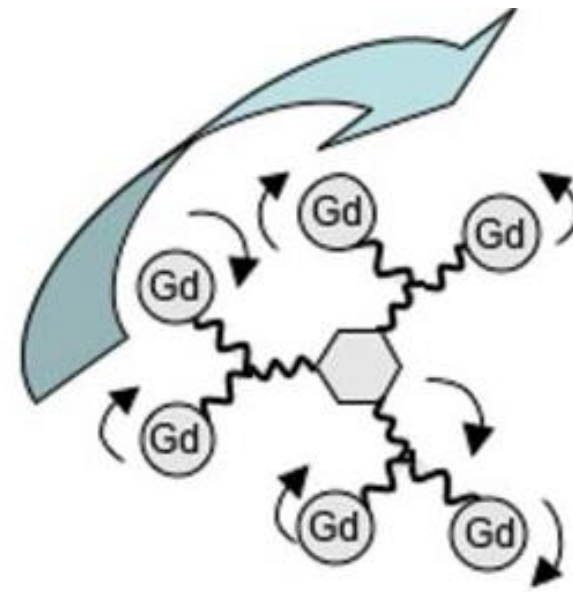
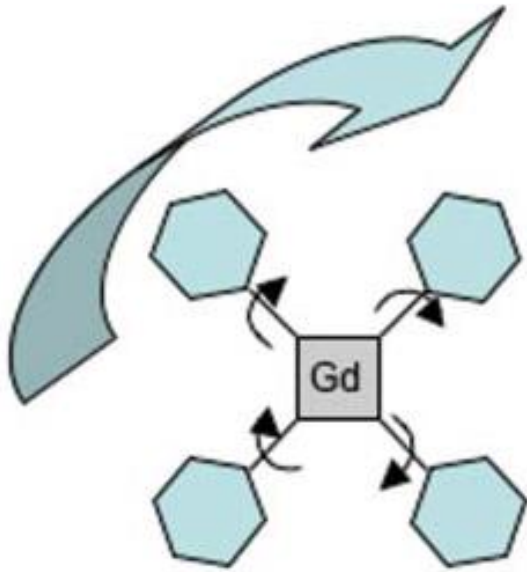


DO3AphenOH

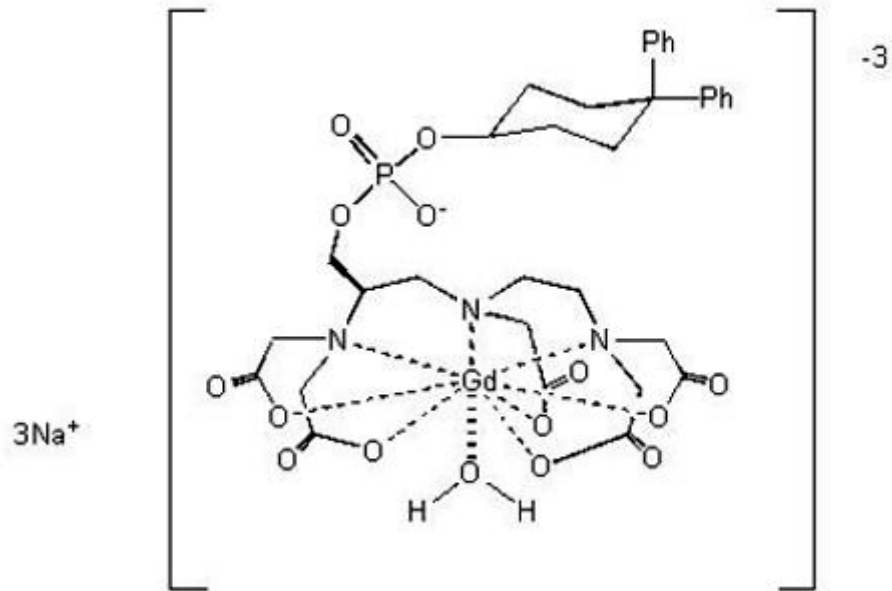
Nuovi leganti polidentati per CA di Gd(III)



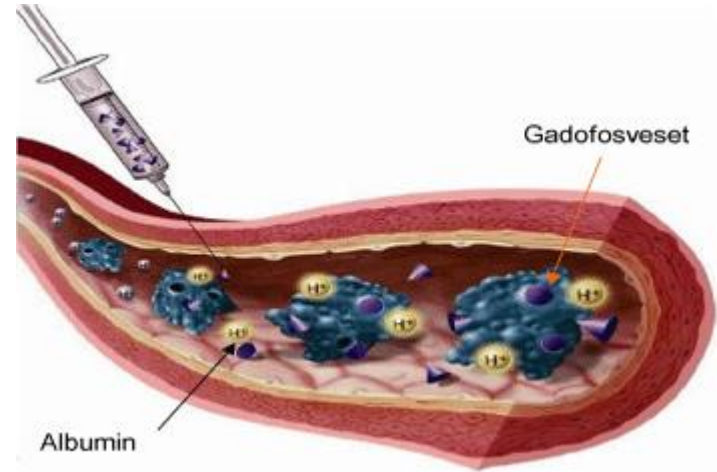
Strategie per aumentare τ_M



Blood pool contrast agents



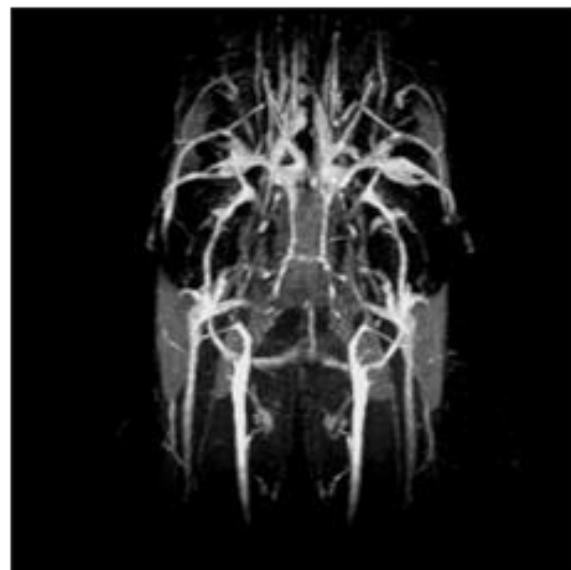
Vasovist[®]



Binding of the C.A. to serum albumin increases its tumbling time (τ_R)



**5 min after
0.1 mmol/kg i.v.
of extracellular CA**



**5 min after
0.015 mmol/kg i.v.
of angiographic ca**

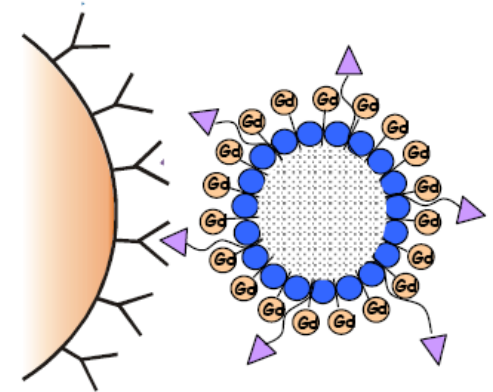
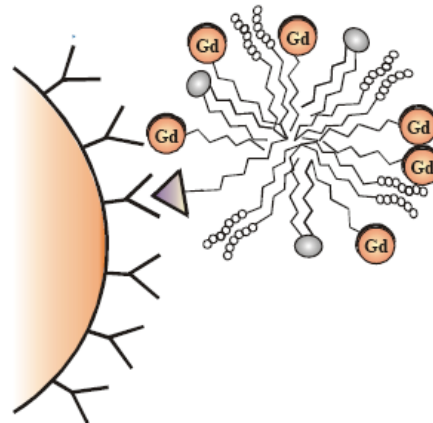
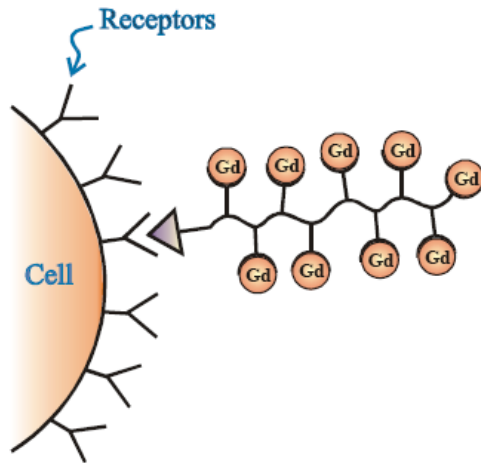
Towards molecular imaging with MRI

The very low concentration of the target requires the delivery of a high number, and possibly efficient, Gd(III) centres

$C > 125 \mu\text{M}$

Several strategies can be adopted

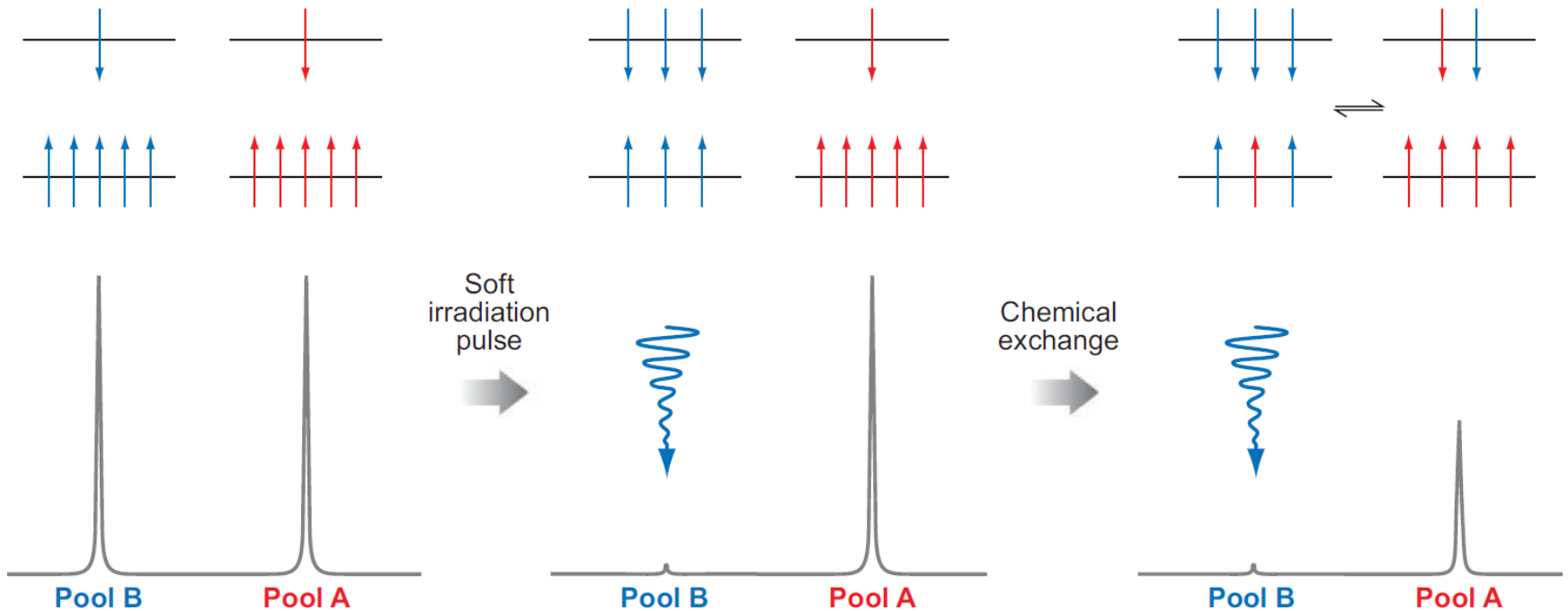
- Gd-chelates covalently or non-covalently linked to biocompatible polymer (proteins, polysaccharides, etc...)
- Self-assembling of complexes (e.g. micelles)
- Use of Gd-loaded nanoparticles (e.g. liposomes,...)



CEST Contrast Agents

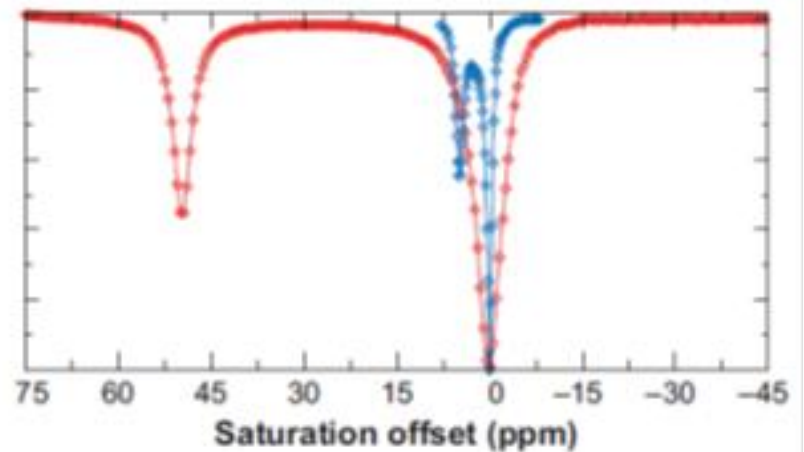
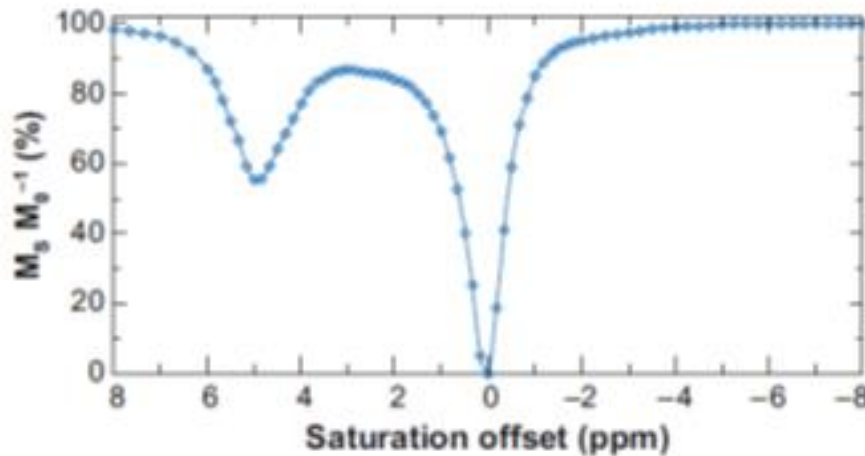
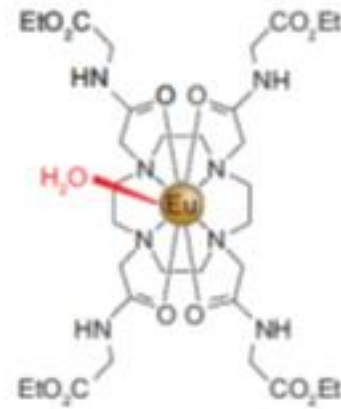
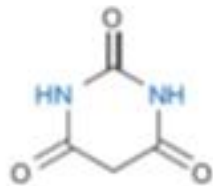
Chemical Exchange Saturation Transfer

composti mobili con protoni in scambio lento con l'acqua di *bulk*



$$k_{\text{CEST}} < \Delta\omega$$

CEST and PARACEST agents: saturation offset



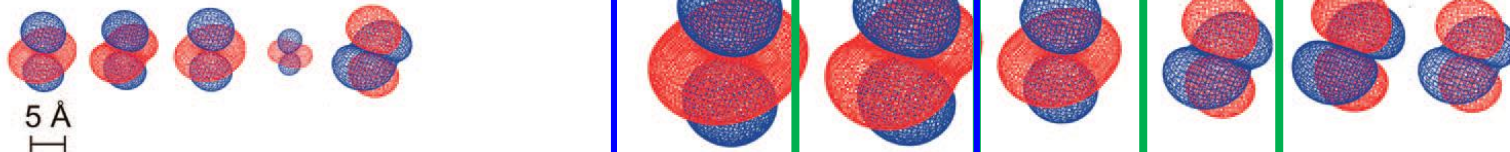
isotropo

Ln	La ³⁺	Ce ³⁺	Pr ³⁺	Nd ³⁺	Sm ³⁺	Eu ³⁺	Gd ³⁺	Tb ³⁺	Dy ³⁺	Ho ³⁺	Er ³⁺	Tm ³⁺	Yb ³⁺	Lu ³⁺
$\mu_{\text{eff}}/\mu\text{B cald.}$	0.00	2.54	3.58	3.68	0.85	0.00	7.94	9.72	10.63	10.60	9.59	7.57	4.53	0.00
$\chi/10^{-32}\text{m}^3$	0.0	5.6	11.2	11.4	0.6	6.0	55.1	82.7	99.2	98.5	80.3	50.0	18.0	0
$\Delta\chi_{\text{ax}}/10^{-32}\text{m}^3$	0	2.1	3.4	1.7	0.2	-2.3	0	42.1	34.7	18.5	-11.6	-21.9	-8.3	0
$\Delta\chi_{\text{rh}}/10^{-32}\text{m}^3$	0	0.7	2.1	0.4	-0.1	-1.6	0	11.2	20.3	5.8	-8.6	-20.1	-5.8	0

PRE



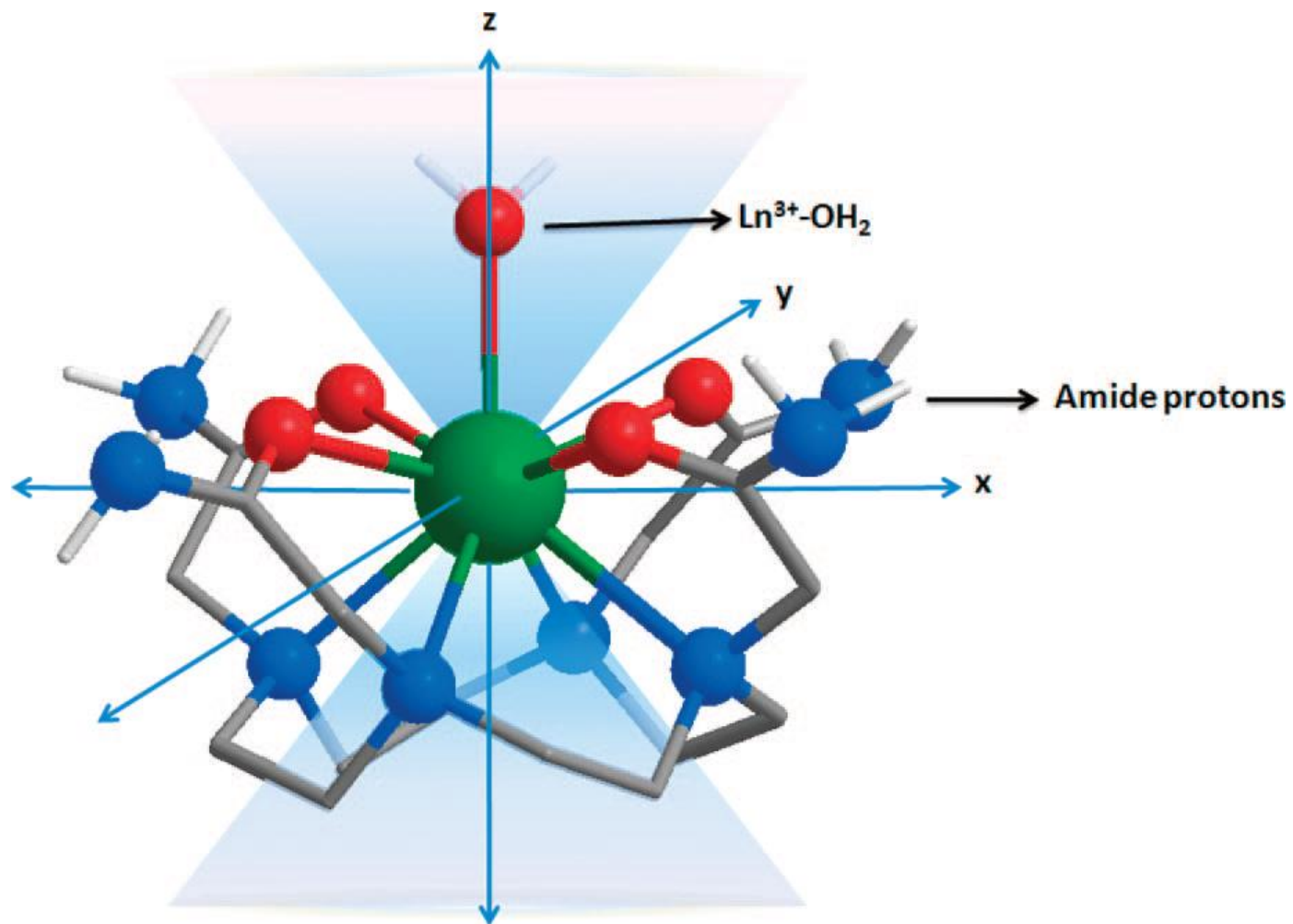
PCS

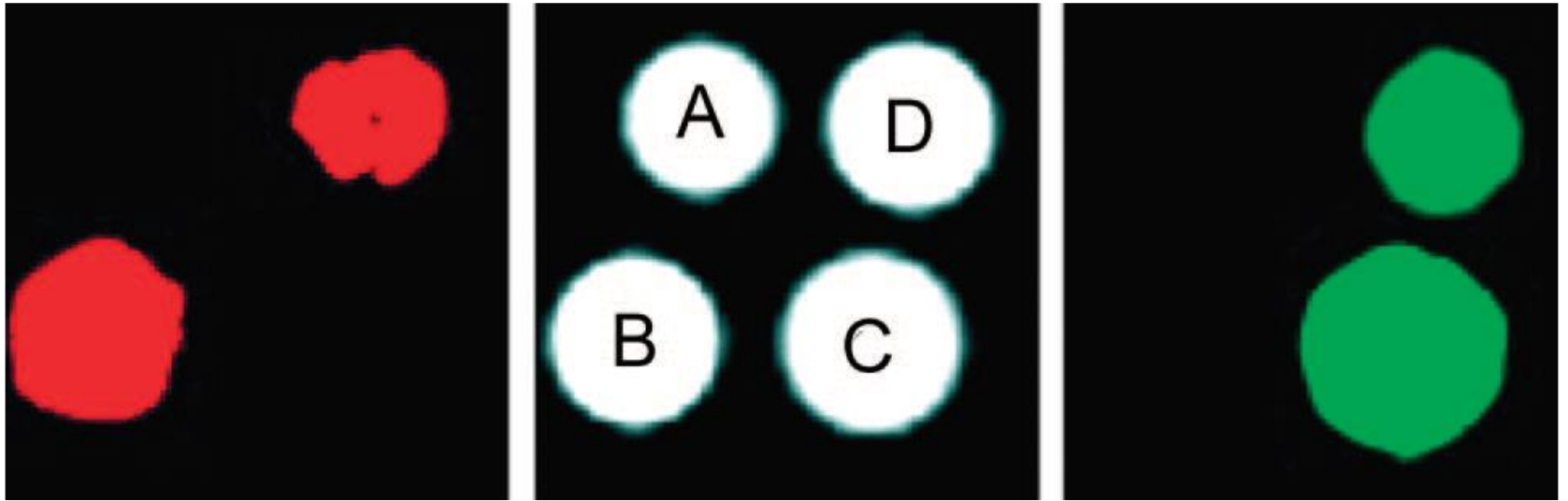


PRE = *Paramagnetic Relaxation Enhancement*

PCS = *Pseudo-Contact Shift*

determina la variazione di chemical shift indotta da ciascuno ione sui nuclei vicini e le iso-superfici rappresentano la grandezza e il segno del chemical shift





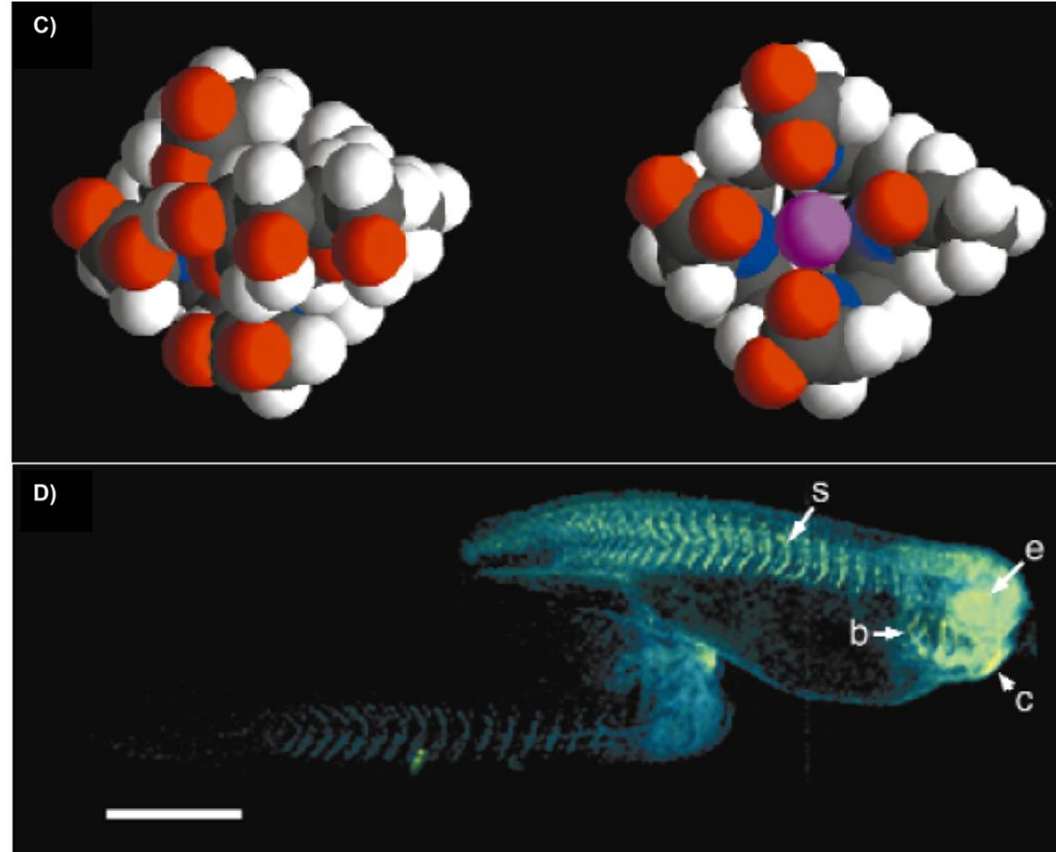
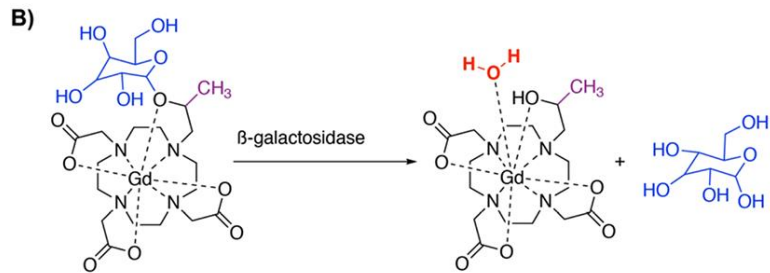
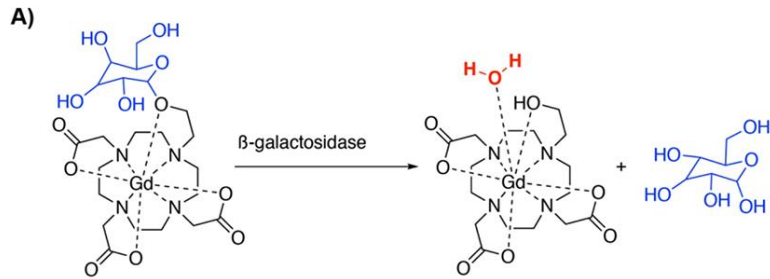
$B = [\text{Tb-DOTAMGly}]^-$

$C = [\text{Eu-DOTAMGly}]^-$

$D = [\text{Tb-DOTAMGly}]^- + [\text{Eu-DOTAMGly}]^-$

Responsive (*smart*) CA

Sensore della β -galattosidasi



55 M water signal was imaged and this signal was augmented by 0.5 mM contrast agent which, in turn, was augmented by a 4 μ M β -galactosidase enzyme concentration (right image)

T₂ contrast agents

super-paramagnetic iron oxide particles (SPIO)

Ø = 60 – 250 nm

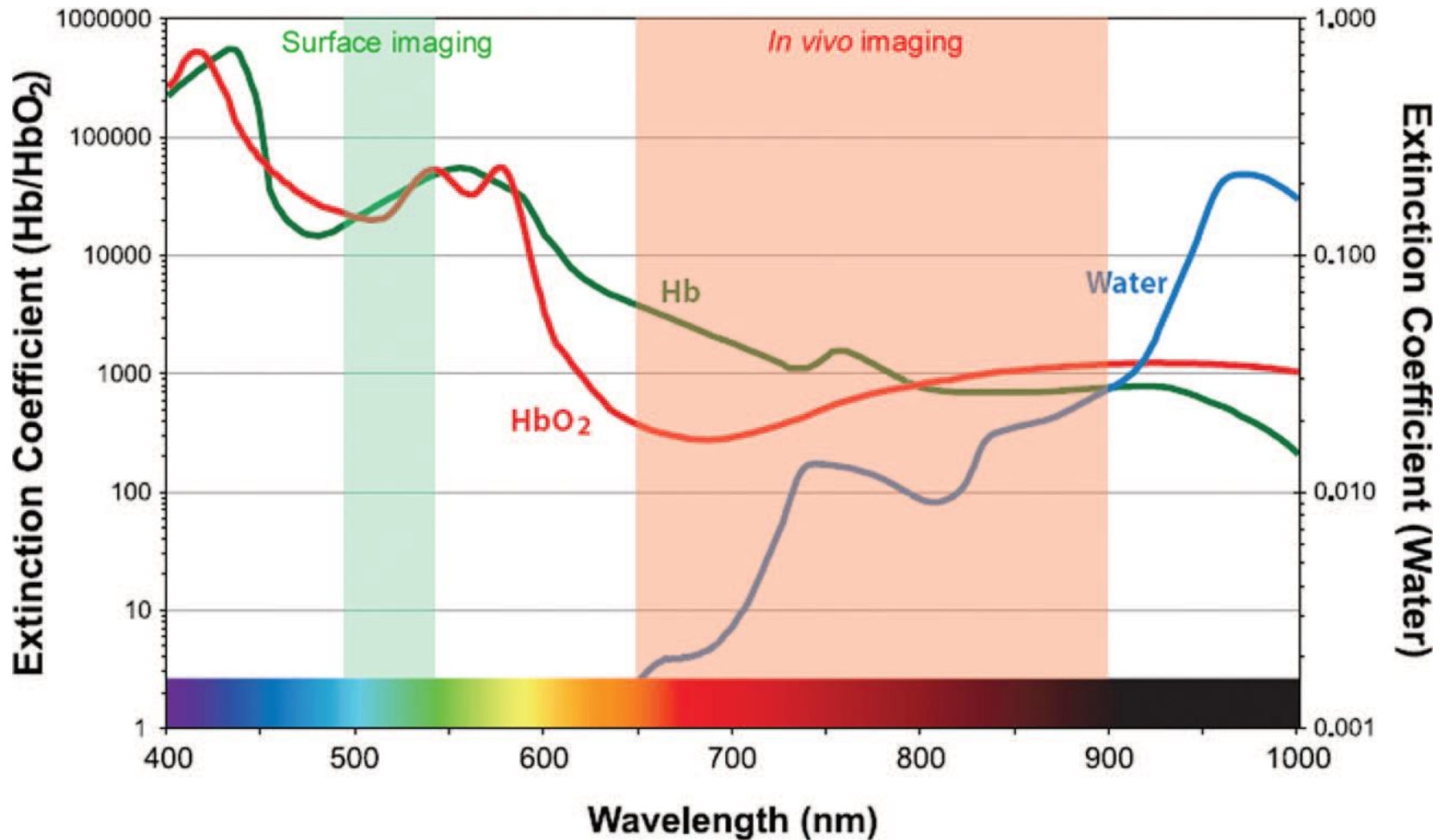
Pre-Clinical Agent	Commercial Name	MR Target	Status
AMI-25	Ferumoxide, Feridex, Endoderm	Liver	Approved
OMP	Abdoscan	Bowel	Approved
AMI-121	Gastromark, Ferumoxsil, Lumirem	Bowel	Approved
SHU555A	Resovist	Liver	Approved (EU, Japan, Australia), Phase III (USA)
AMI-227	Combidex, Sinerem, Ferumoxtran	Lymph Node Metastases	Phase III
CODE 7228	Feraheme, Ferumoxytol	Vasculature	Phase II

Coating biocompatibile: destrano, amido, glicosammino-glicano, silossani organici, copolimeri stirene-divinilbenzene solfonati,....

Imaging ottico

- Sensibilità paragonabile a quella di SPECT e PET
- Possibilità di agenti *switchable (responsive)*
- Possibilità di *time-resolved detection*
- **No quantificazione**

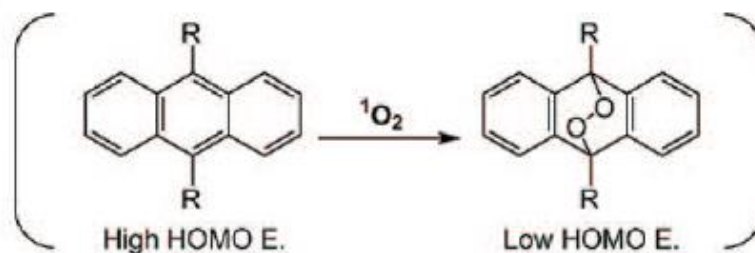
- Window
- Stokes shift
- Brightness
- Stability



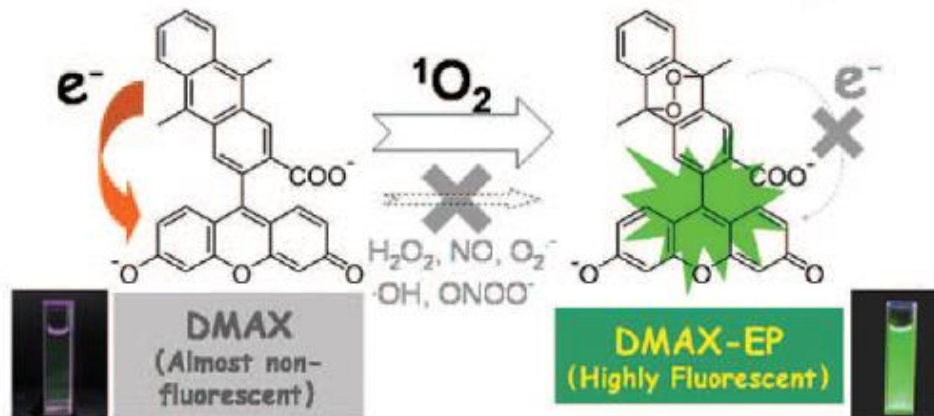
Esempio di *switchable fluorescent probe* sensore di $^1\text{O}_2$

(a) Singlet Oxygen Probes

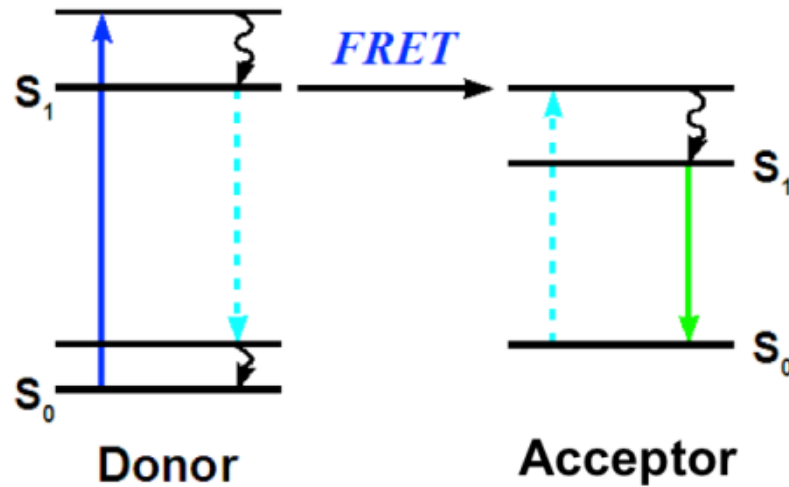
Key reaction: Endoperoxide formation



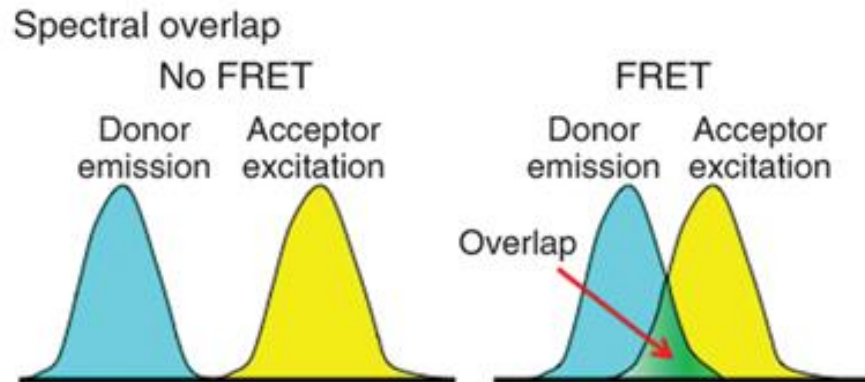
Reaction scheme for detection of singlet oxygen



FRET fluorescence – resonance energy transfer



$$1/r^6$$

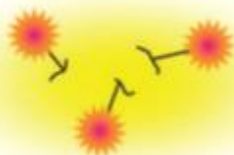


a) Self-quench (Homo-FRET)



Weak fluorescence

dequench



Strong fluorescence

b) Fluorophore protein interaction



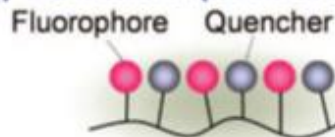
Weak fluorescence

dequench



Strong fluorescence

c) Quencher (Hetero-FRET)

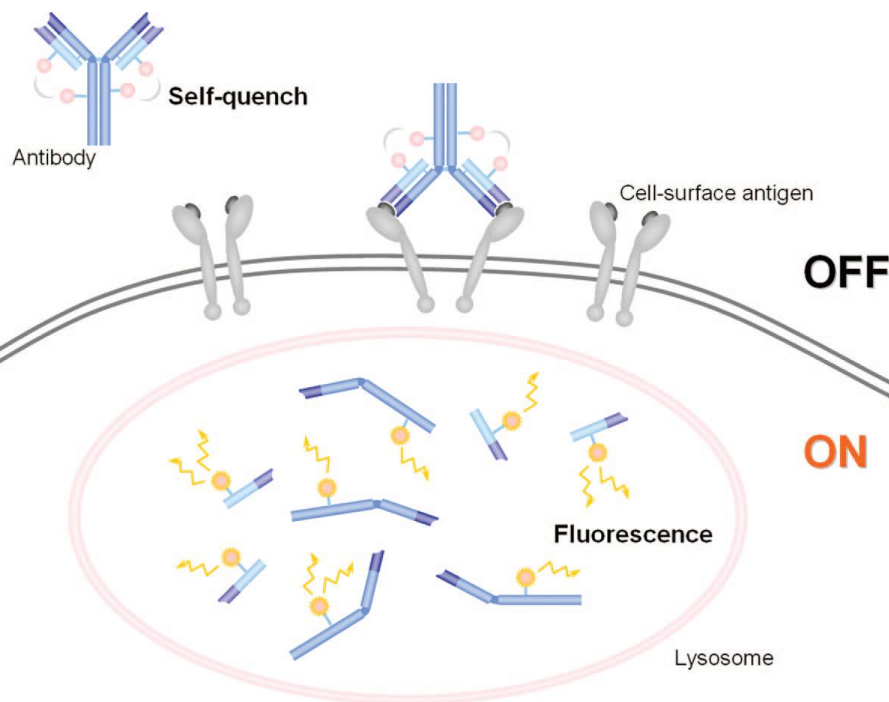
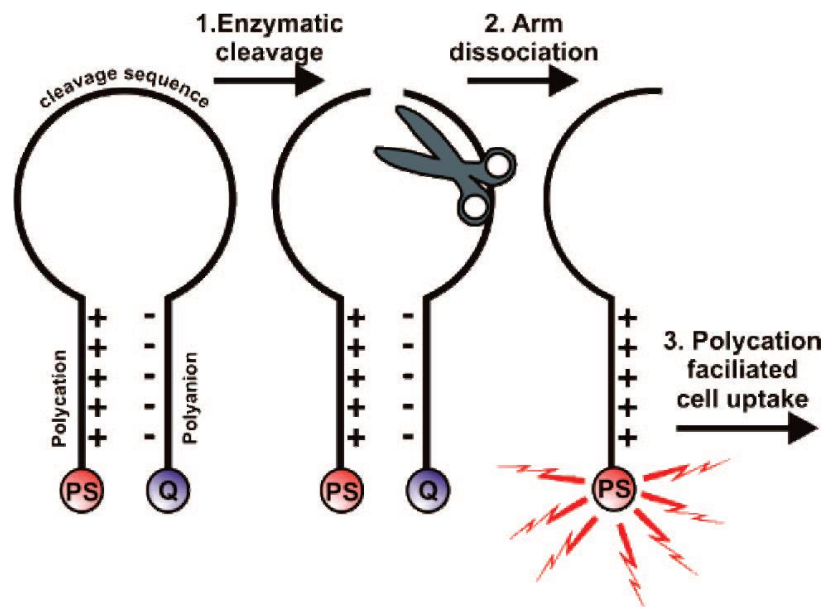


Minimal fluorescence

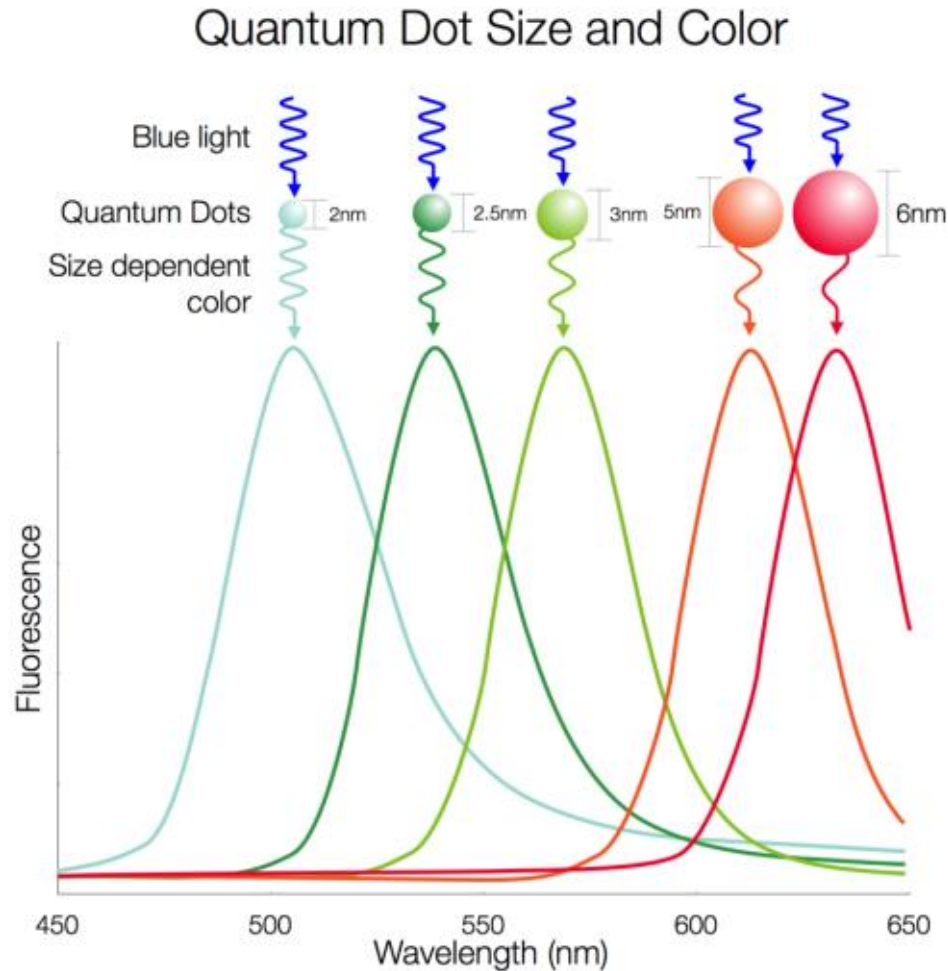
dequench



Strong fluorescence



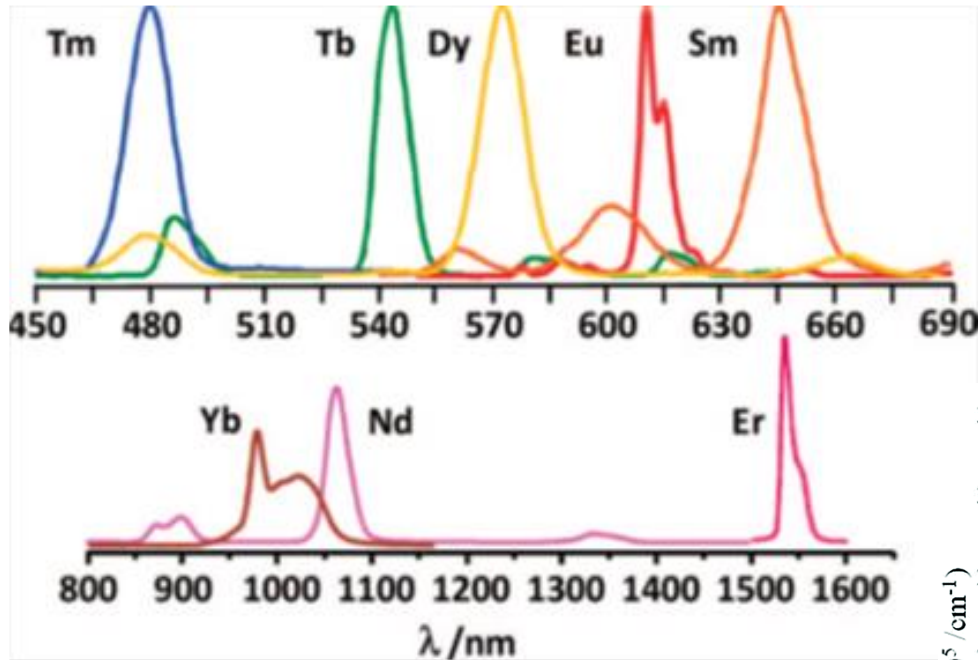
quantum dots (QD) nano-cristalli di semiconduttori (e.g. CdSe)



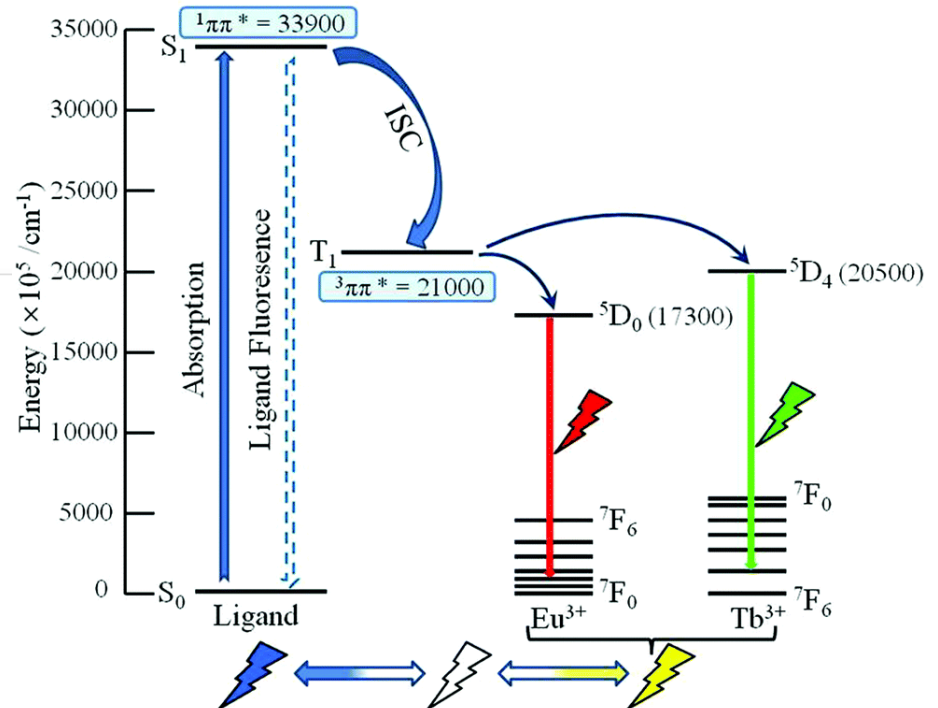
Ø 2 – 10 nm

Banda di emissione stretta, molto intensa e modulabile con le dimensioni del QD

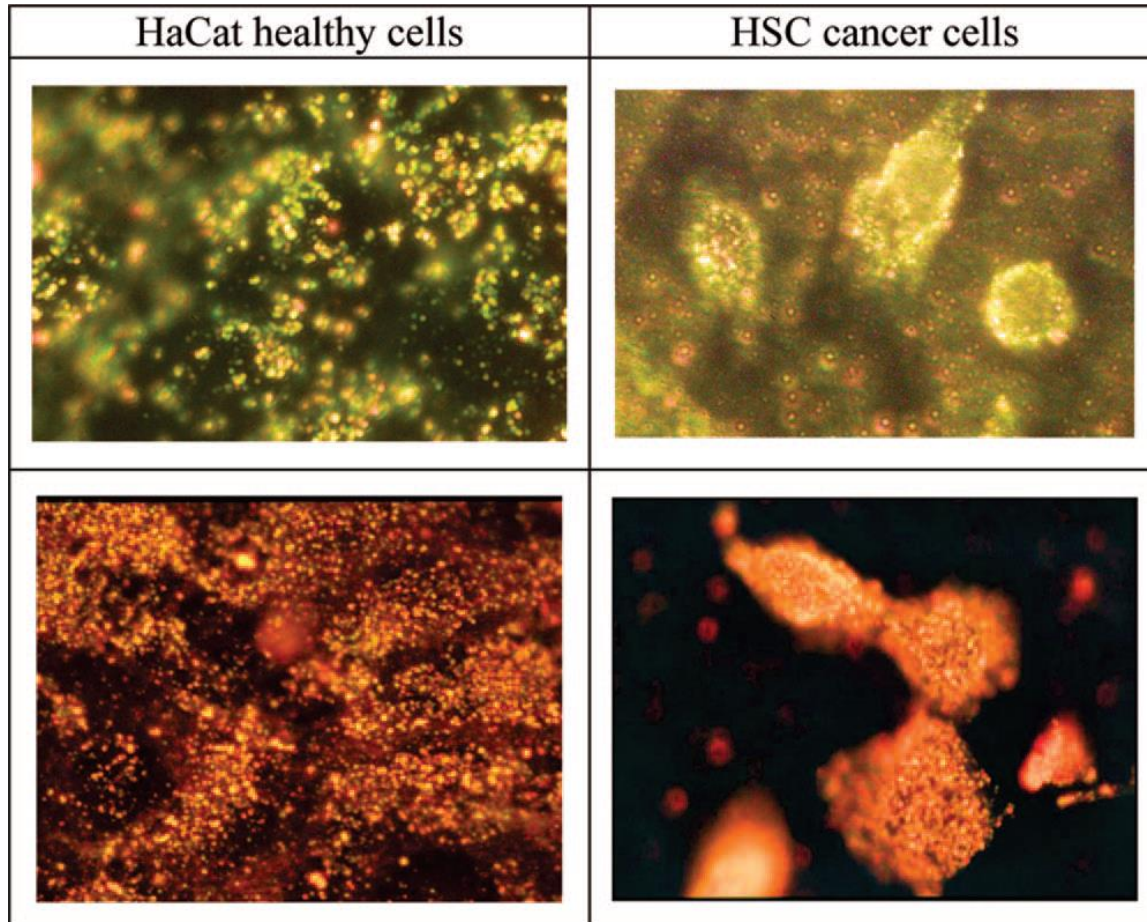
Complessi dei lantanidi



Effetto antenna dei leganti



Dark-field fluorescence imaging con AuNP



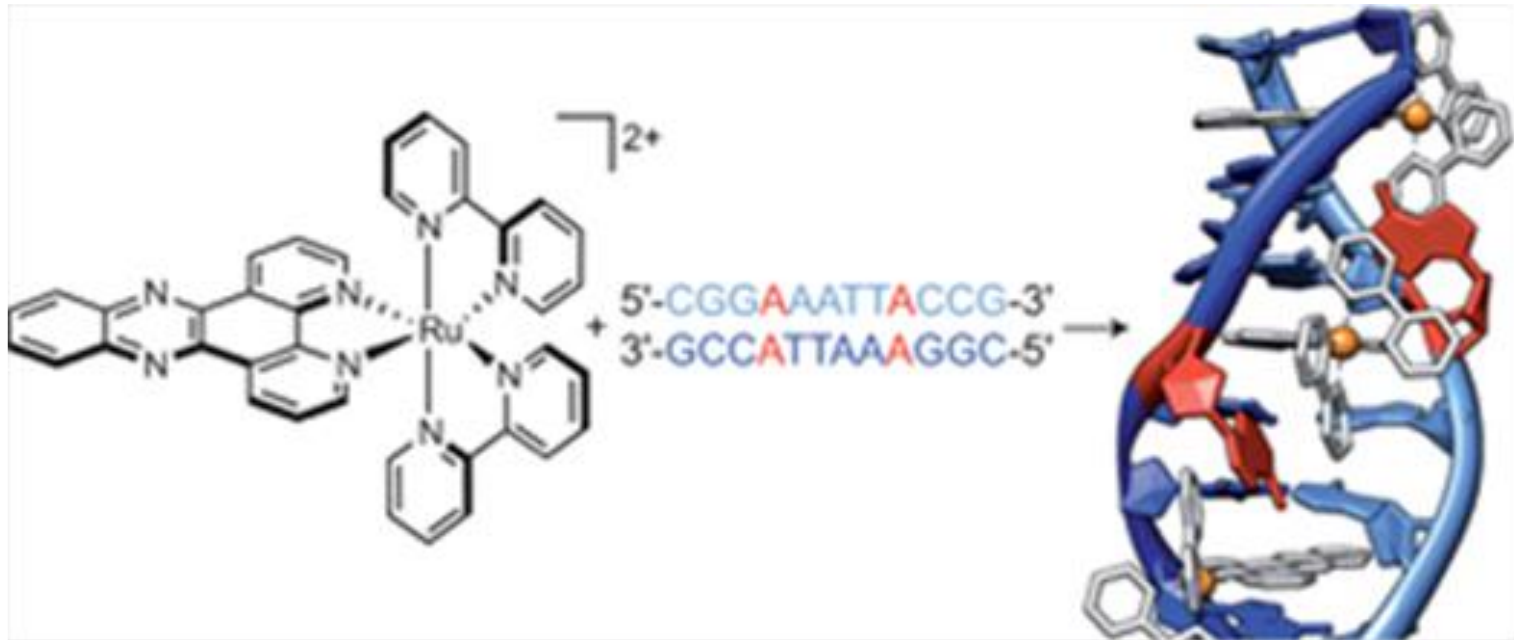
Au nanospheres

Au nanorods

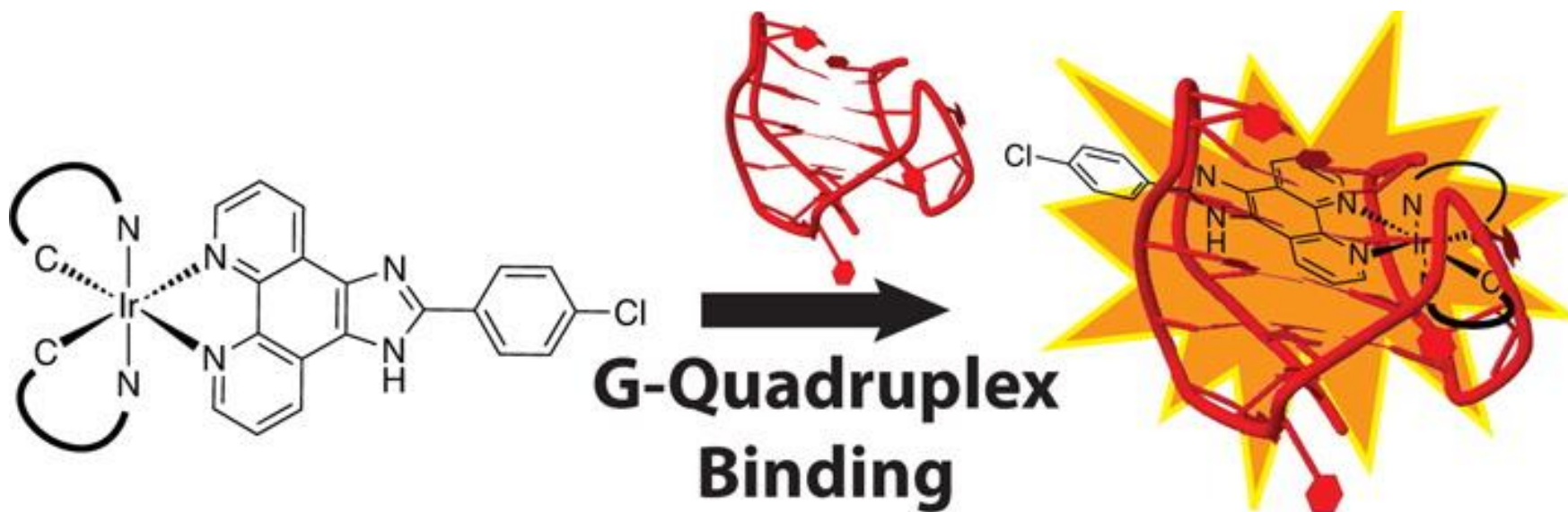
AuNP coniugate a anticorpi anti-EGFR

EGFR = *epidermal growth factor receptor*, marcatore tumorale

Complessi polipiridilici di Ru(II) come *DNA light switch*



G-quadruplex sensing



Sviluppi futuri

Multimodal imaging agents and theranostics

