In-vivo imaging with luminescent nanoparticles emitting in the second biological window

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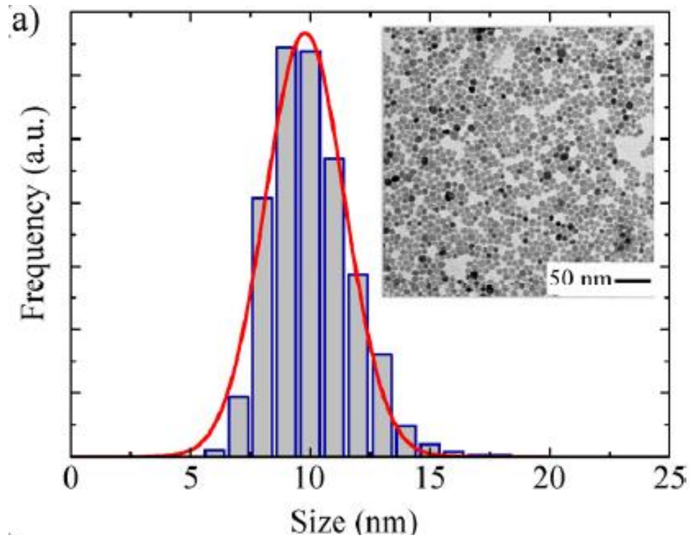
Fluorescence bio-imaging

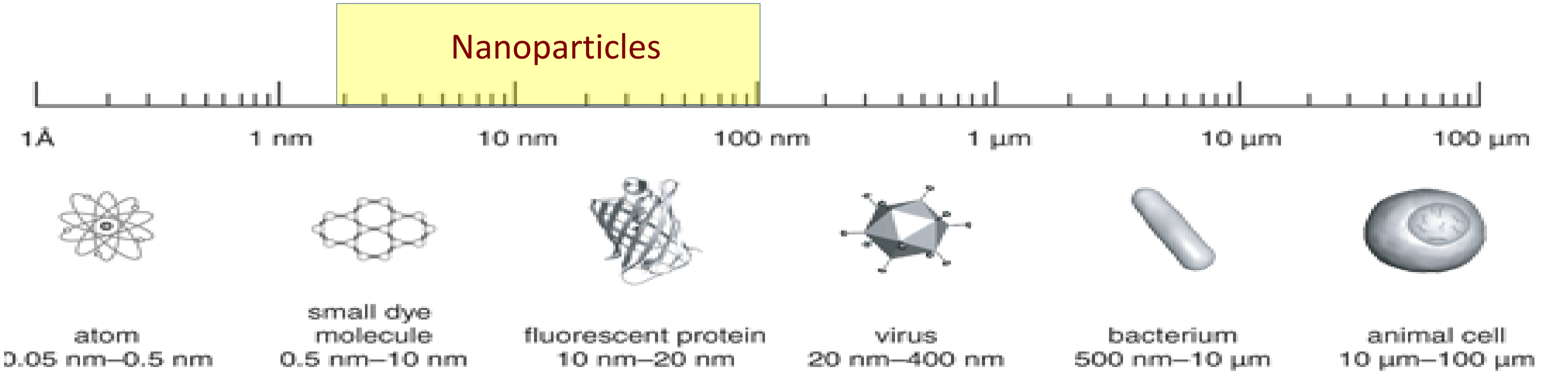
Efficient and biocompatible fluorescent nano-probes for "in vivo" biomedical imaging with high spatial resolution, fast feedback, and large photoluminescence quantum yield



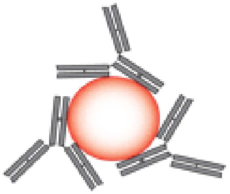
NANOTECHNOLOGY- Metal, semiconductor, and insulating inorganic systems, as well as polymeric compounds

Nanoparticles in medicine: Nanomedicine

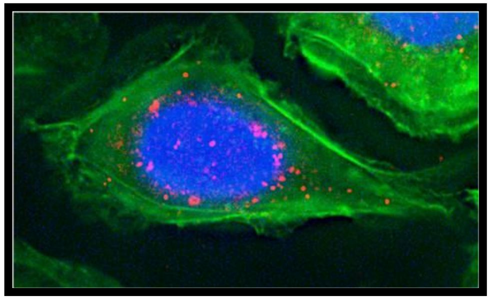
Why nanoparticles?



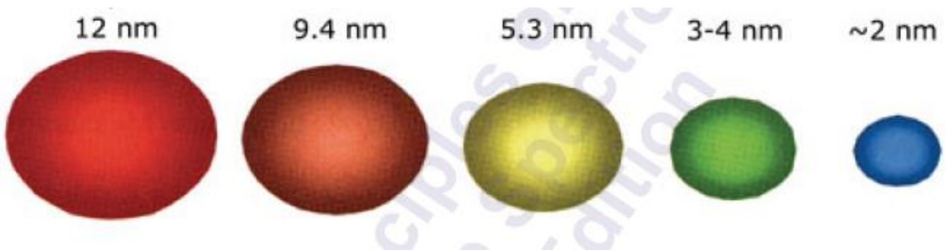
-Large surface to accommodate functional groups (diagnosis, therapeutic…)



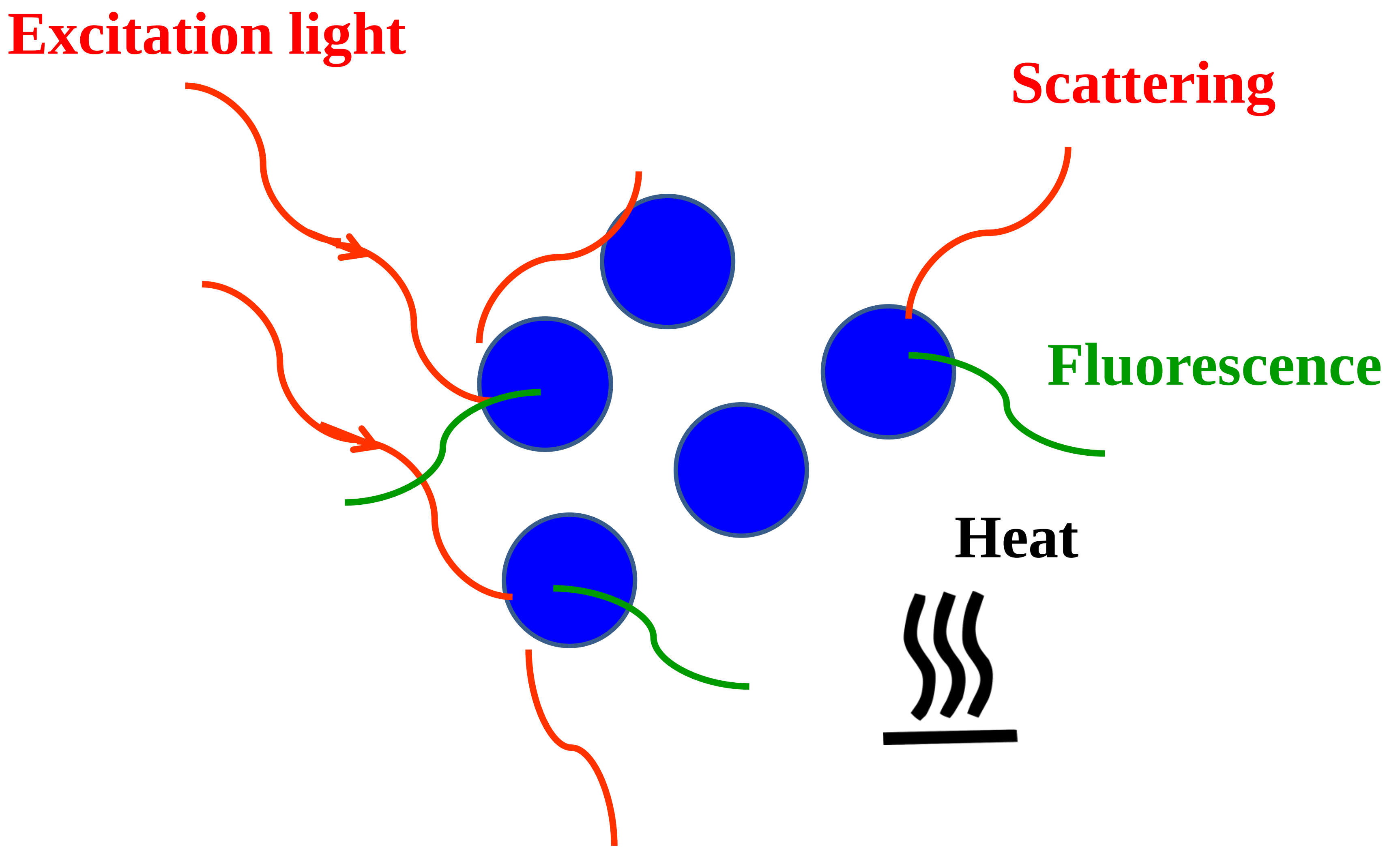
- Interact in a singular way with biomolecules, proteins and can be up-taken by cells.



- Spectral properties (semiconductors and metals) depend on the particle size.



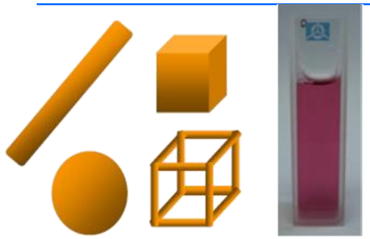
Light excitation of Nanoparticles



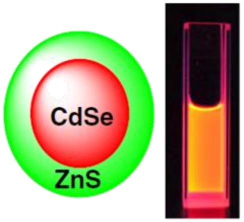
Scattering - Fluorescence (imaging and therapy); Heat (therapy)

Nanoparticles for multiphoton excitation

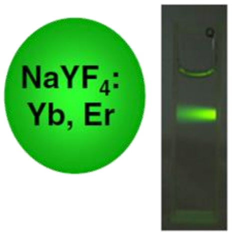
Gold Nanoparticles	• Nanorods, Nanocages, Nanospheres…
Quantum Dots	• CdS, CdSe, ZnS, ZnSe, PbSe, PbS
Rare earth activated insulating crystals	• $NaGdF_4$, $NaYF_4$, CaF_2, • SrF_2



Gold Nanostructures

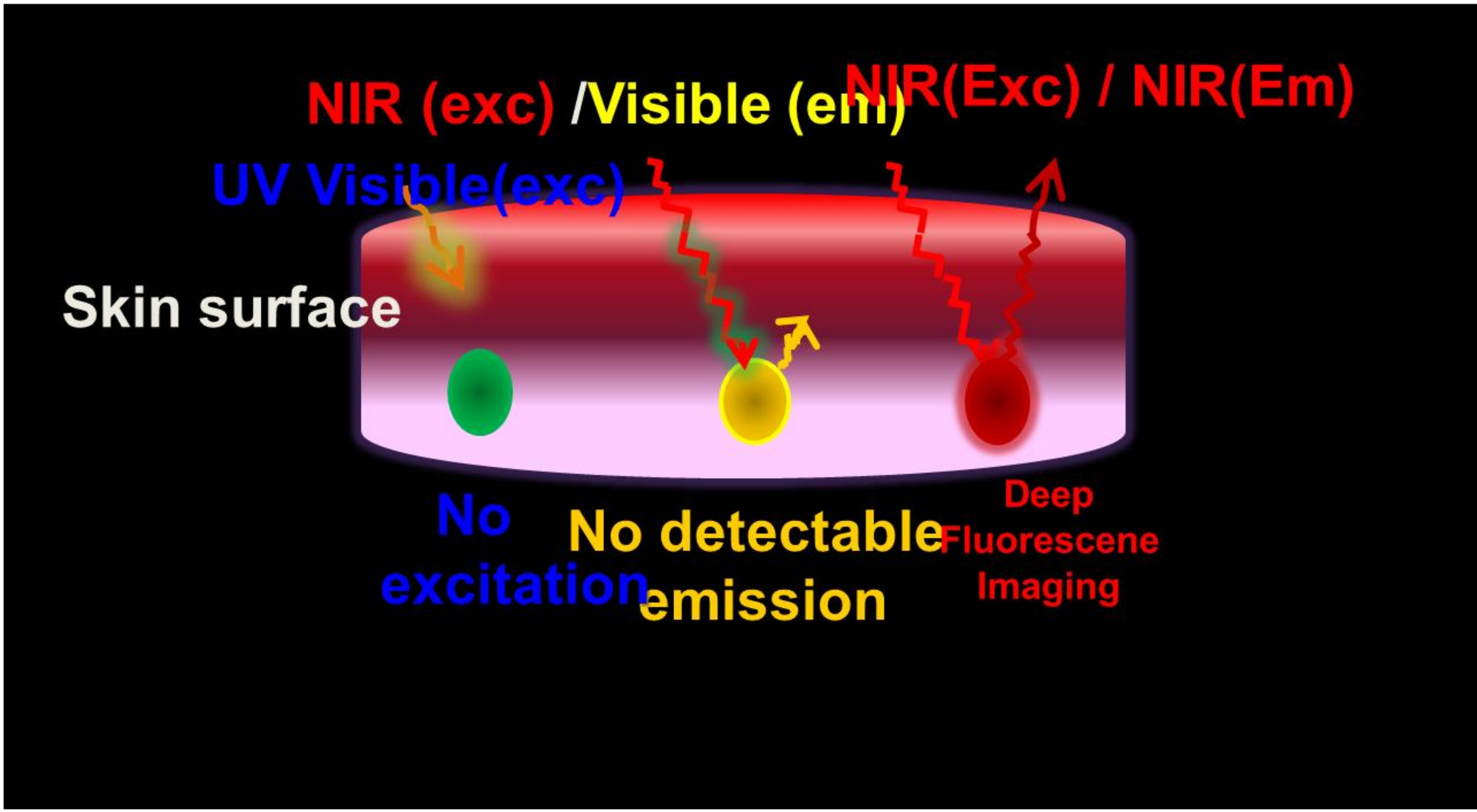


Quantum Dots

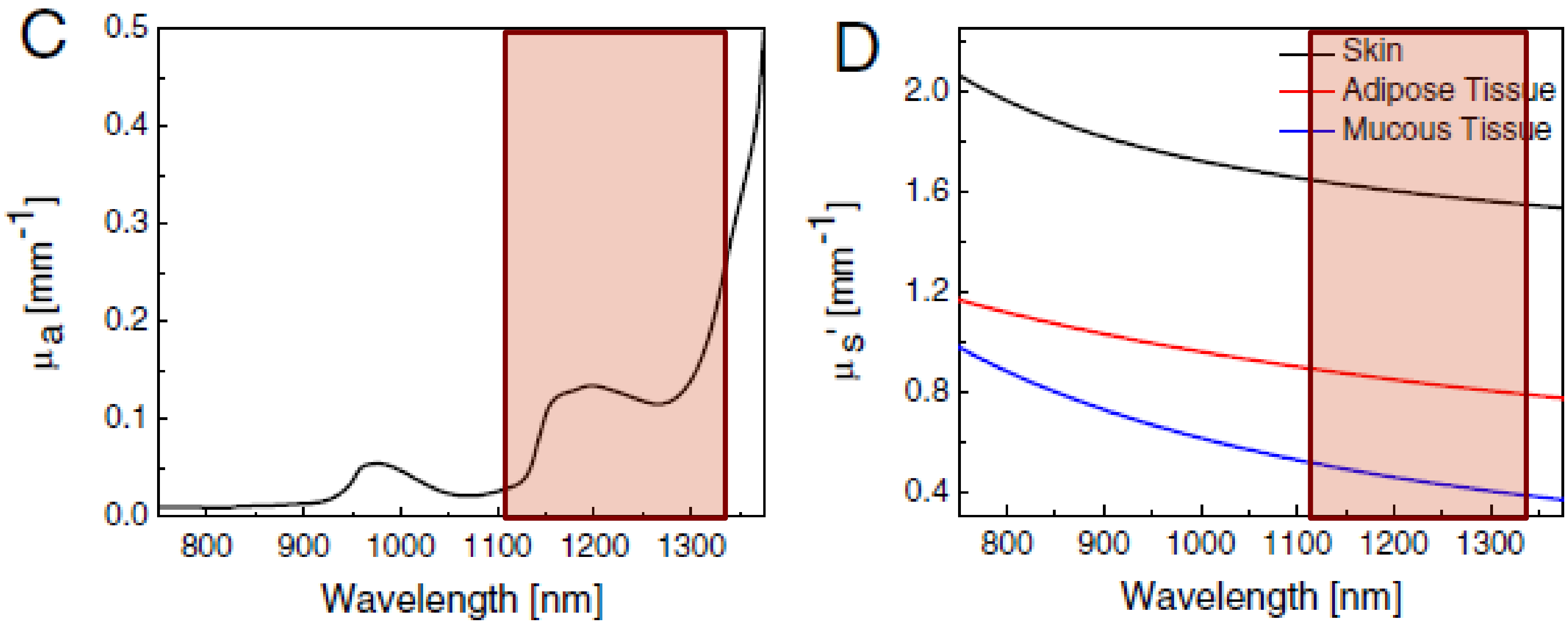


Up-conversion Nanoparticles

-Towards "In vivo imaging": Novel IR-IR probes.



SECOND BIOLOGICAL WINDOW

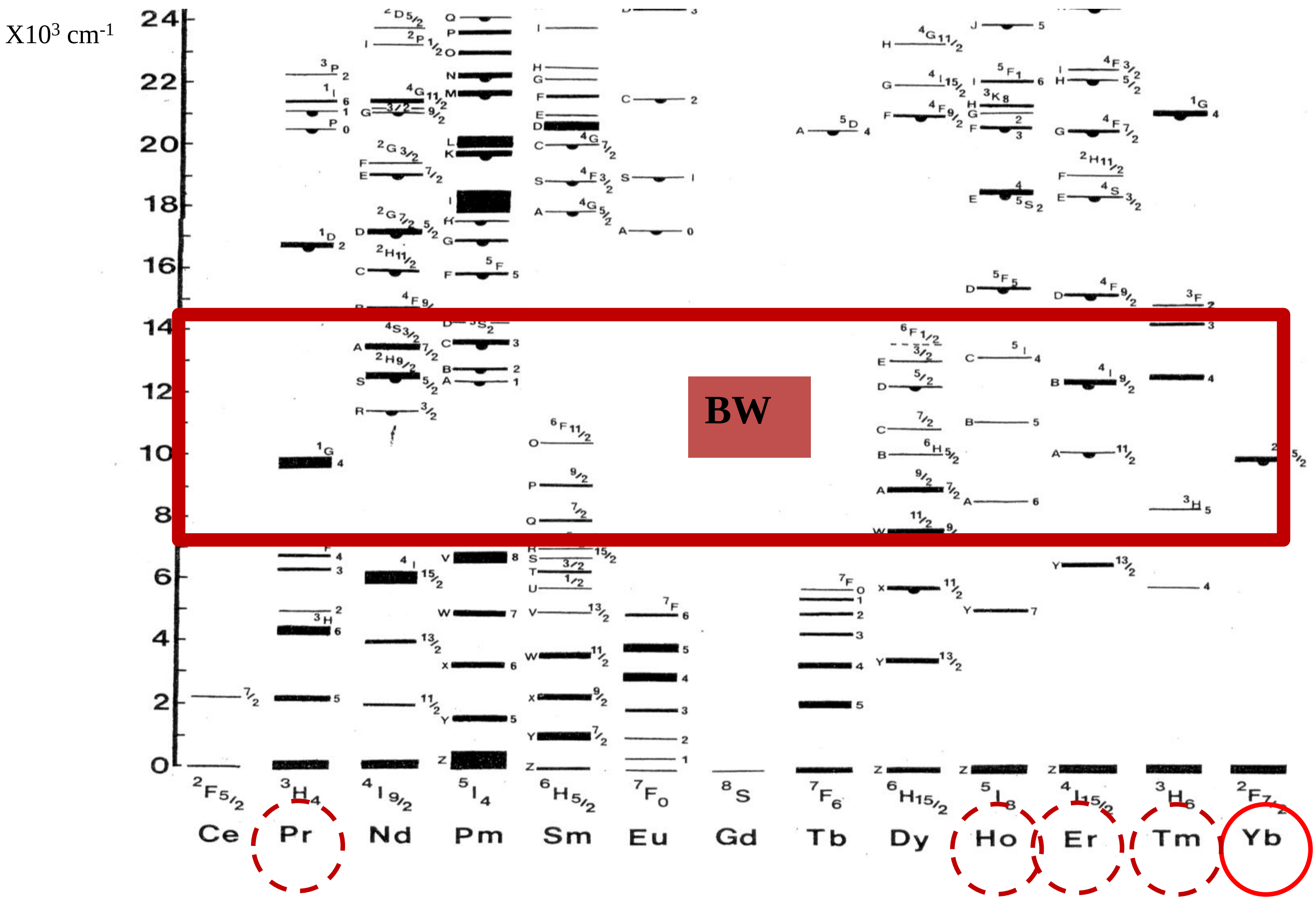


ABSORPTION **SCATTERING**

K. Welsher at al.PNAS | May 31, 2011 | vol. 108 | no. 22 | 8943

The irruption of rare earth ions for deep tissue imaging

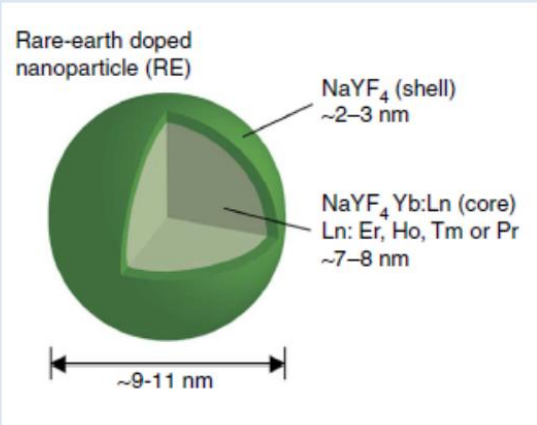
Multiemission possibilities

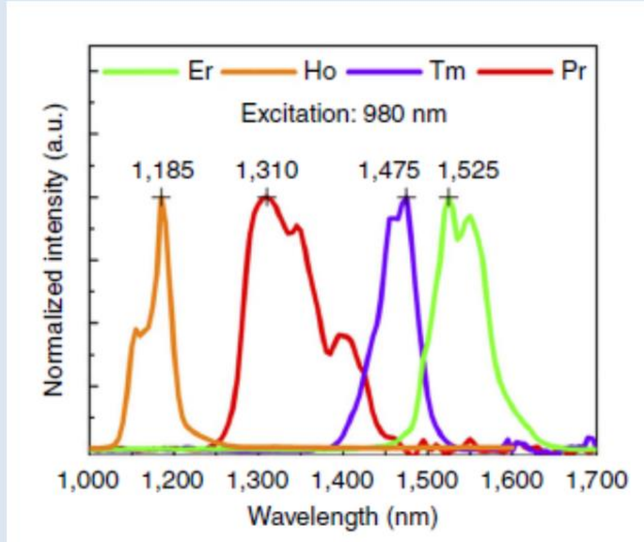


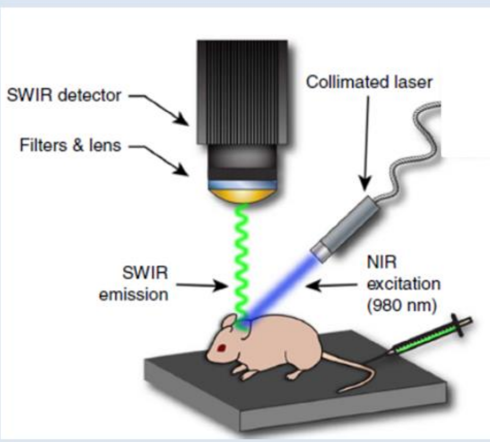
The irruption of Rare earth ions for deep tissue imaging

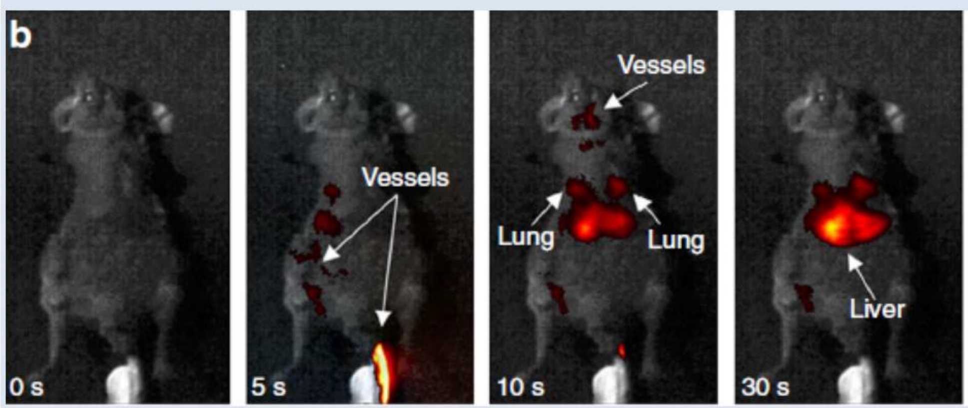
2013.- Real time infrared *in vivo* imaging by rare earth doped nanoparticles.

D.J. Naczynskie et al. Nature Communications 4, 2199 (2013)



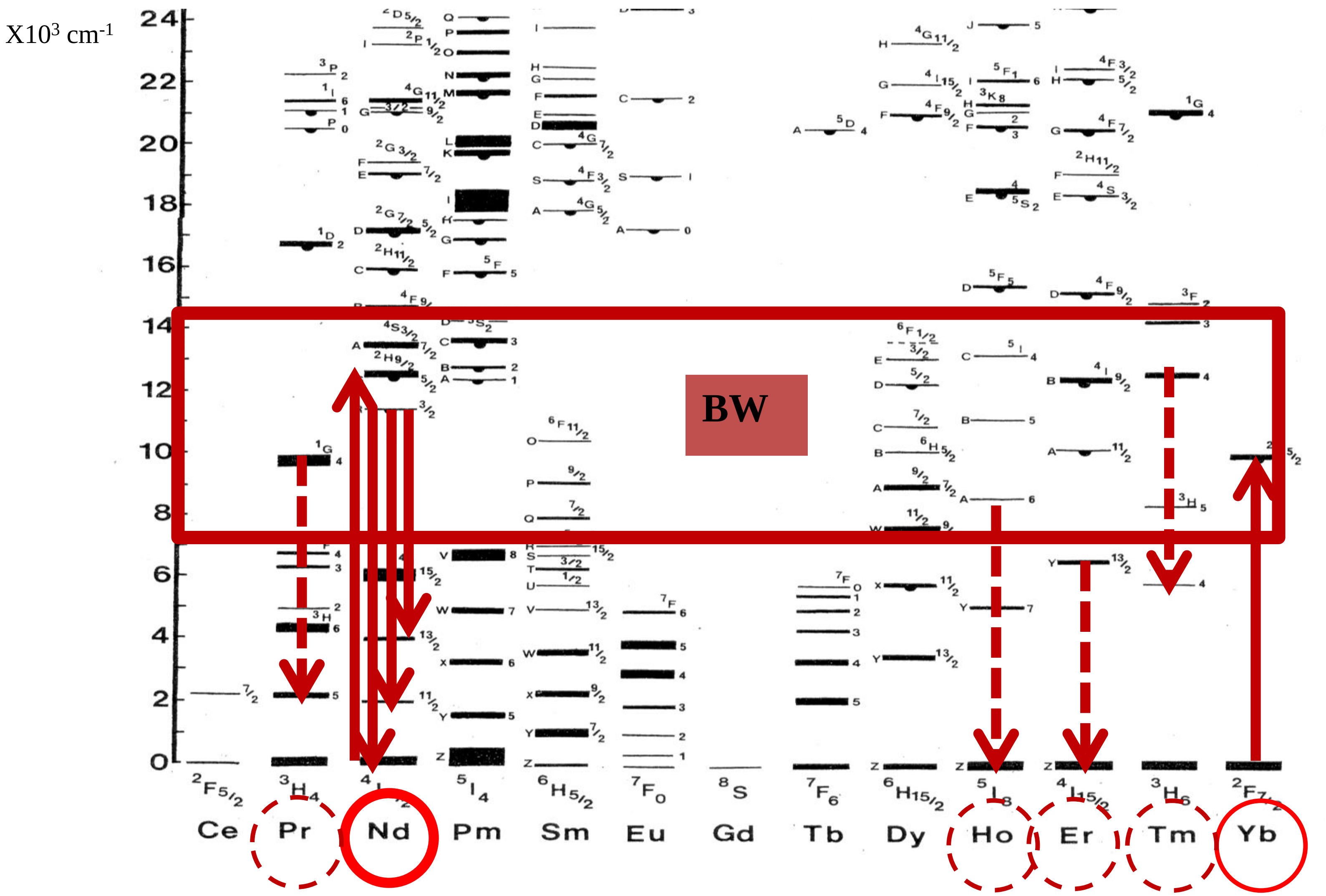




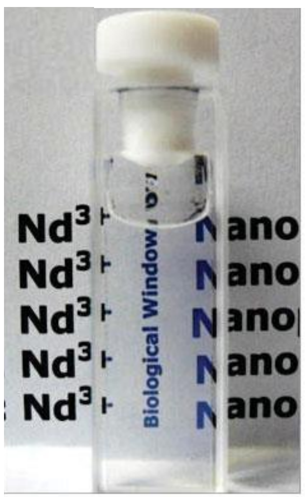


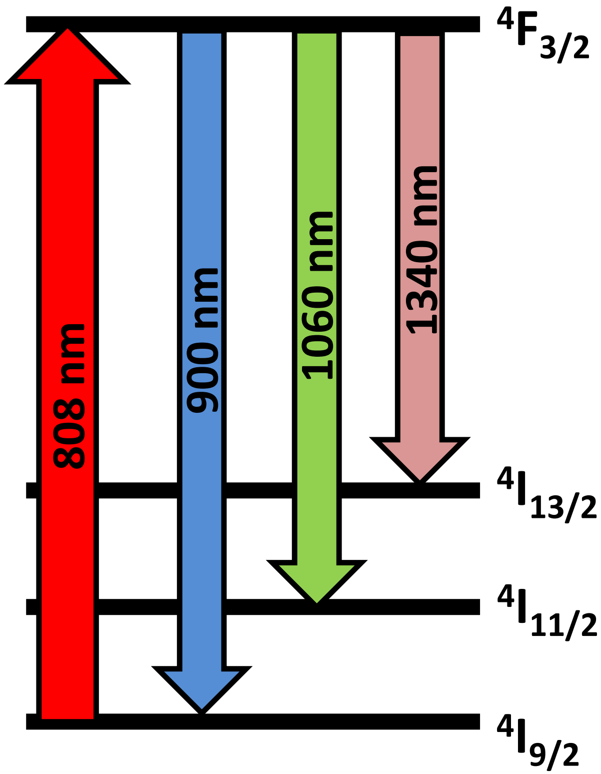
Circulation time: over tens of hours

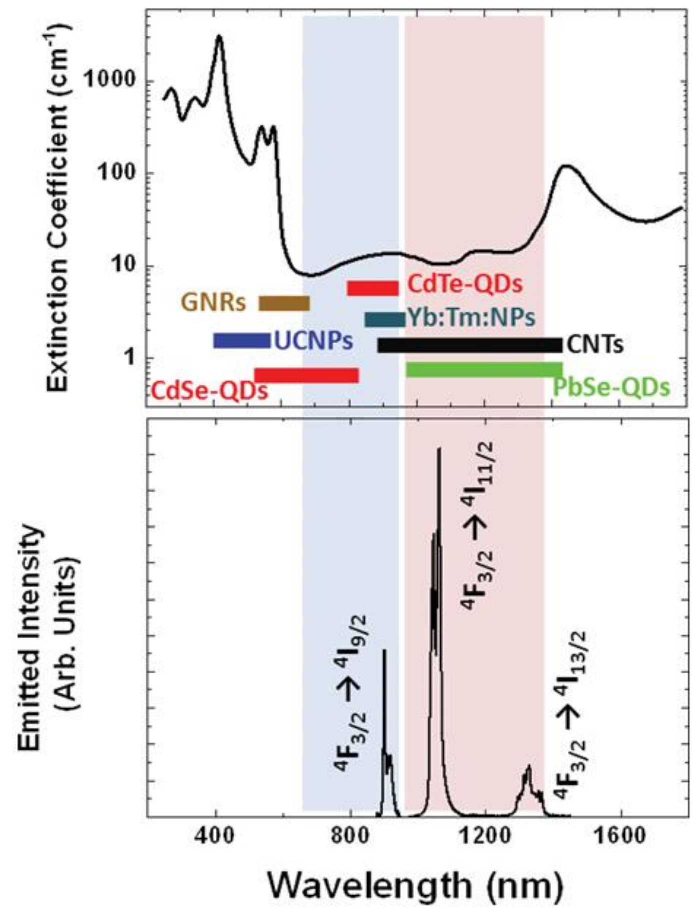
Nd^{3+} ion for deep tissue imaging



Nd^{3+} doped materials

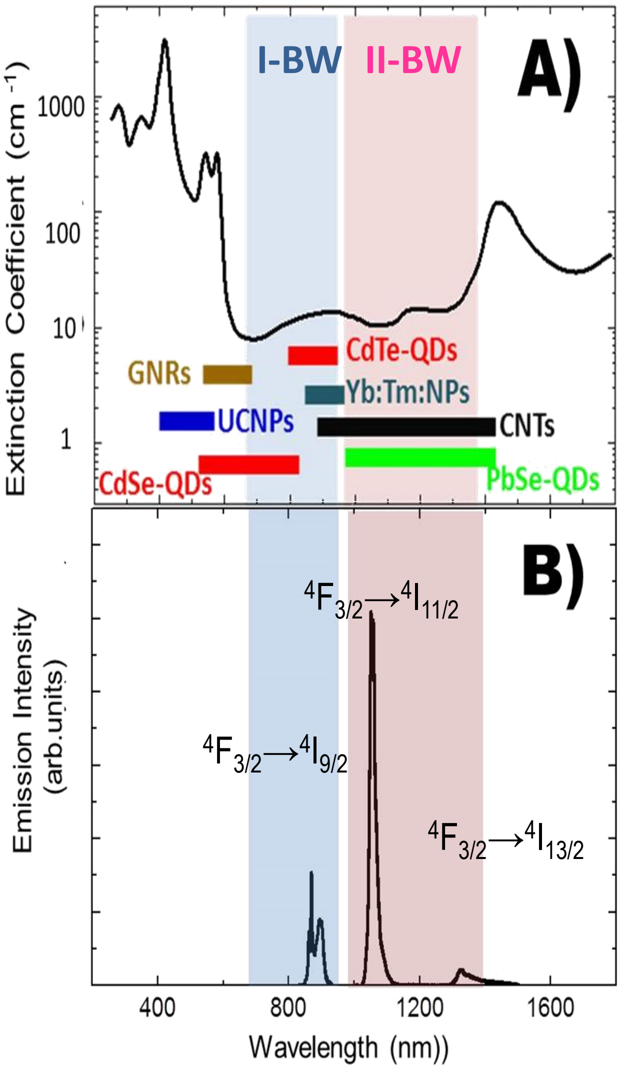






Nd^{3+} ion works at all the transparency windows of human tissues …

Issues of tissue bio-imaging:



Attenuation coefficient of human blood

Emission spectrum of a colloidal solution of Nd^{3+}:SrF_2 NPs in water

1. living tissue inhomogeneity scatters light
2. the biological media (water, blood, haemoglobin, melanin and lipids) act as absorbers in the visible range.
3. human tissue is partially transparent, in <u>the first biological window</u> (700-950 nm) and in <u>the second biological window</u> (1000-1500 nm)

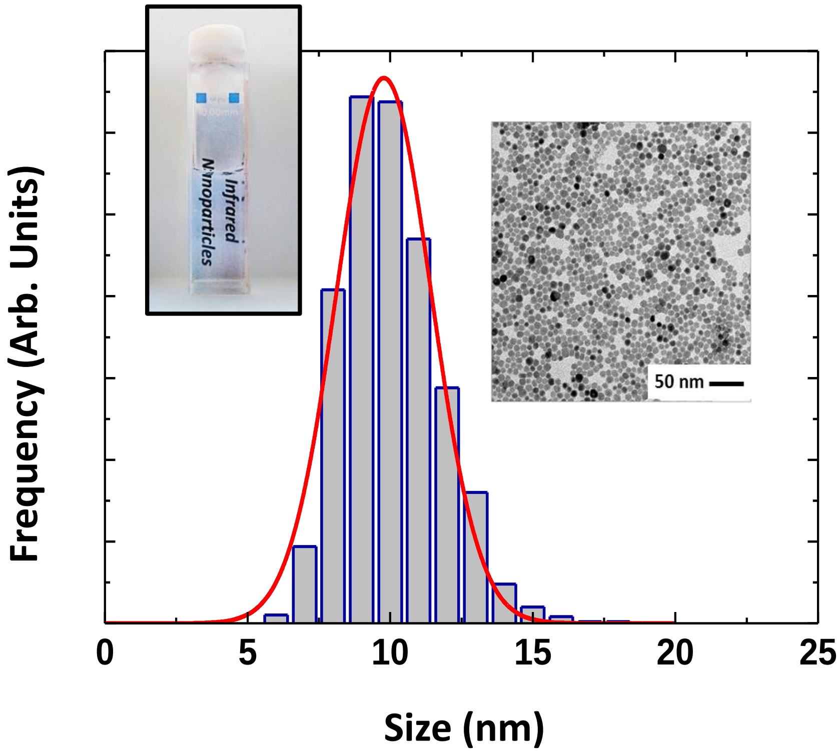
Trivalent lanthanide ion activated fluoride nanocrystals may overcome those problems.

Nd^{3+} ion displays well defined emission bands both in the I-BW and in II-BW that can be efficiently excited at around 800 nm.

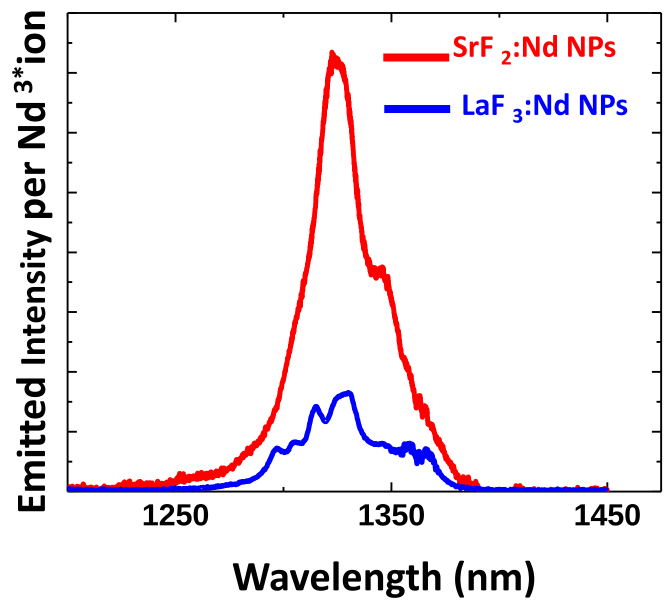
We synthetized and studied a novel type of Nd^{3+}: SrF_2 nanoparticles.

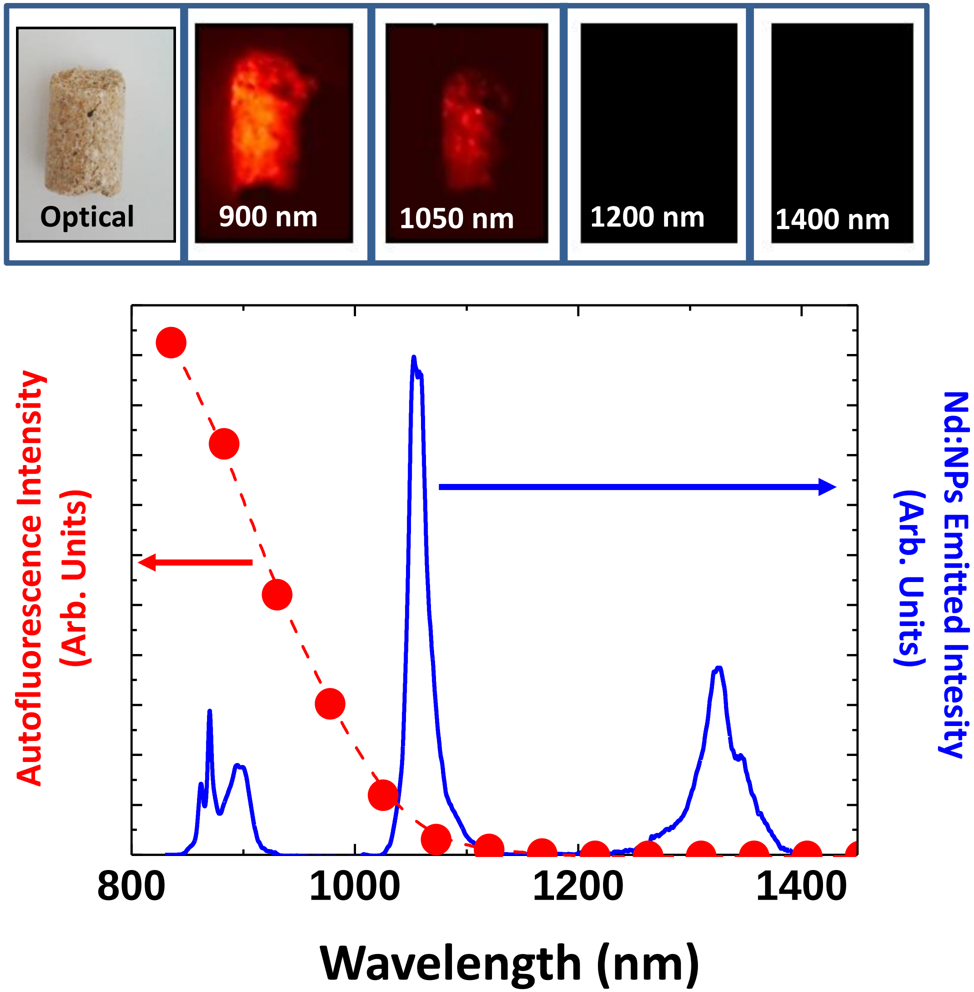
Materials - SrF_2:Nd NPs

- Aqueous colloidal solution of SrF_2 :Nd NPs
- NPs were well dispersed in water as we can notice from the transparency of the solution.
- TEM image reveals monodispersed 3mol% Nd:SrF_2 nanocrystals with a diameter of about 10 nm. No agglomeration is evident.



Due to local charge compensation required when Nd^{3+} is incorporated in a divalent fluoride, its luminescence is almost 10 times enhanced in the divalent SrF_2 NPs with respect to the most recently studied matrix, LaF_3.

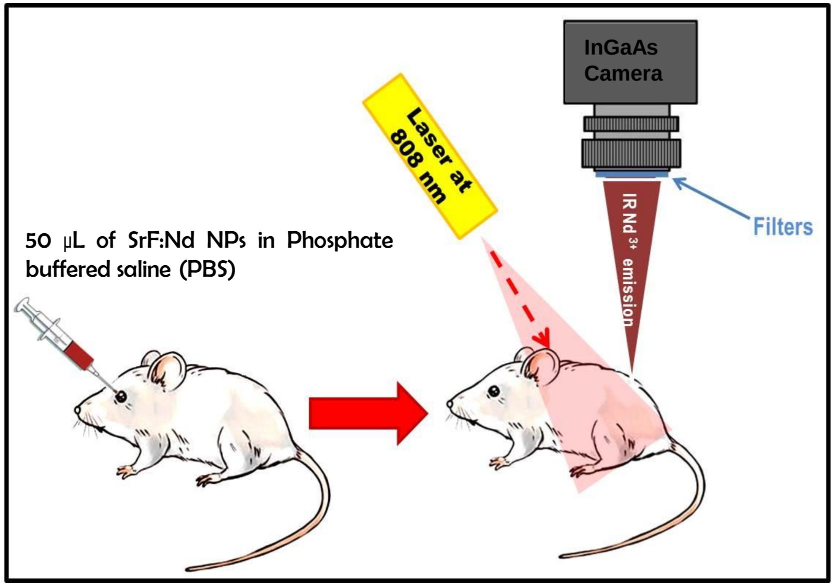


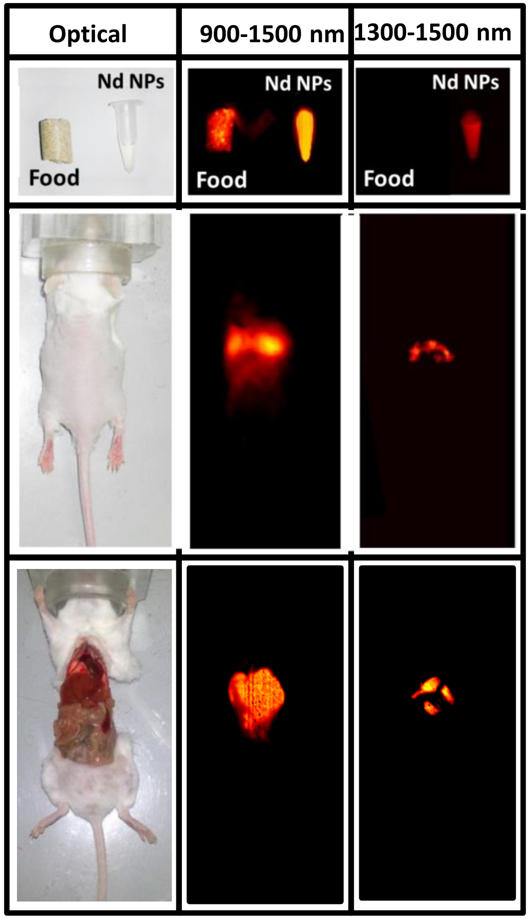


Top.- Optical image of a mouse food pellet and fluorescence images of the same pellet as obtained using different long-pass filters when excited with a 808 nm laser.

Bottom.- Wavelength dependence of the infrared auto-fluorescence generated by a mouse food pellet. Dots are experimental data and dashed line is a guide for the eyes. Bottom graph also includes the room temperature emission spectrum of a colloidal solution of Nd:SrF_2 nanoparticles.

In vivo imaging in the II-BW:

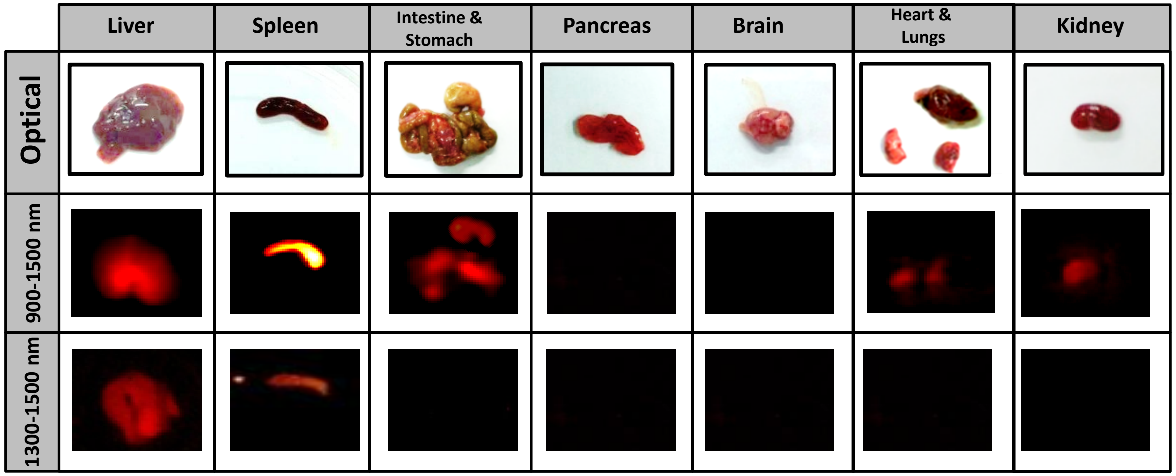


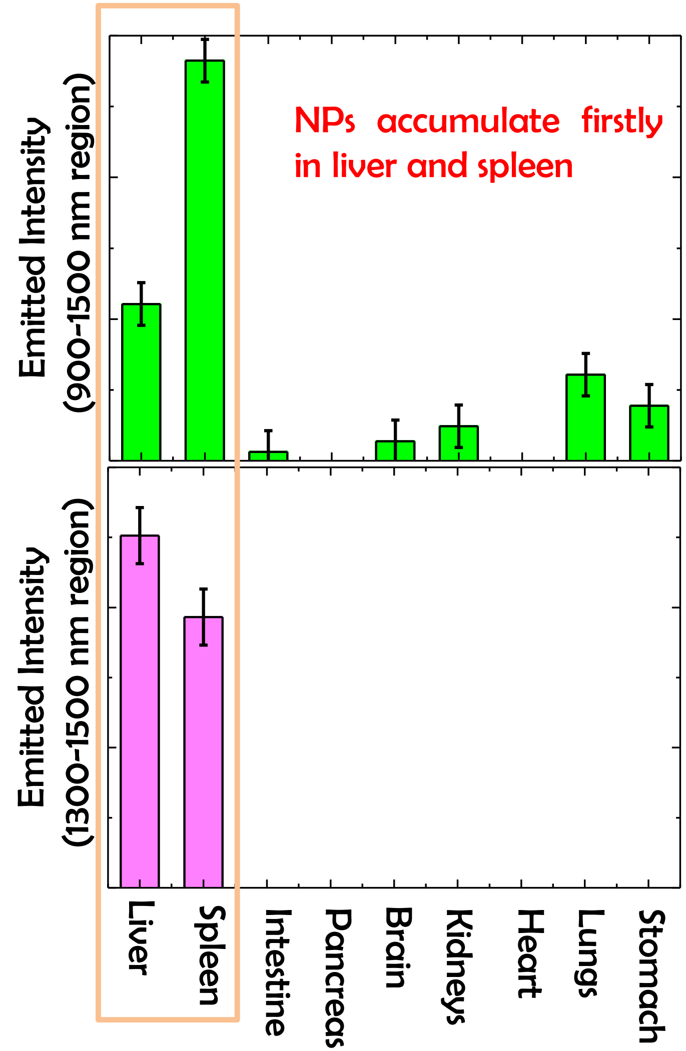


Intensity, shape and resolution change in the IR images, by acquiring the signal in the two different spectral regions.

The *in vivo* fluorescence images reveal that the emission at 1055 nm ion is not the most suitable due to overlap with food luminescence to fed the mice. The Nd^{3+} luminescence at 1330 nm, not affected by the food light output, must be selected.

Biodistribution experiments:

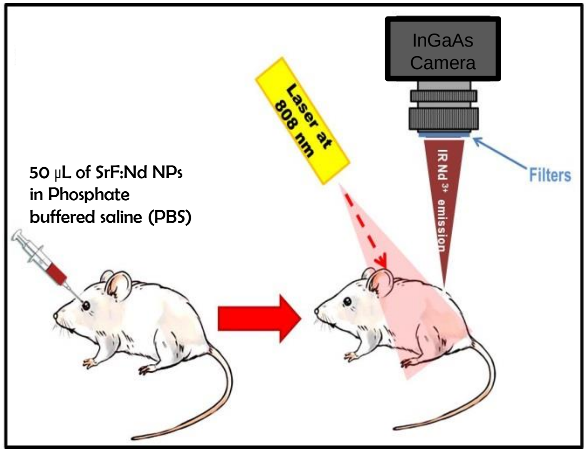


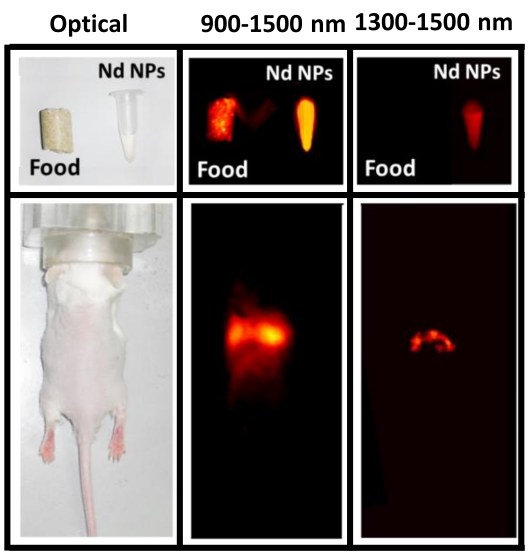


Left- Optical images and fluorescence images in the 900-1500 and 1300-1500 nm spectral detection ranges of the organs extracted from a sacrified mouse after 4 hours of an intravenous injection of SrF_2:Nd NPs.

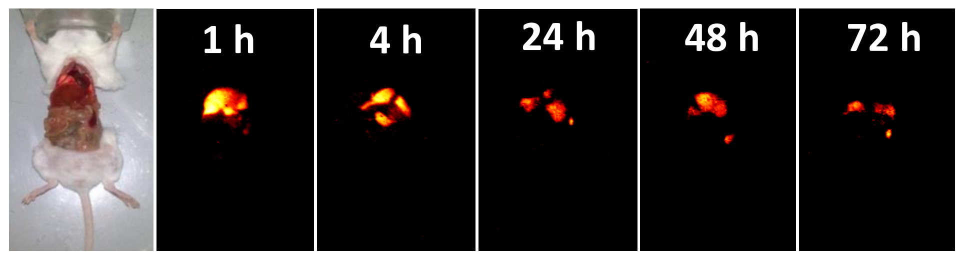
Right- Integrated fluorescence intensity obtained from the different organs in the two spectral ranges (900-1500 nm and 1300-1500 nm). In all the cases, the integrated fluorescence intensity has been normalized by the organ´s weigth.

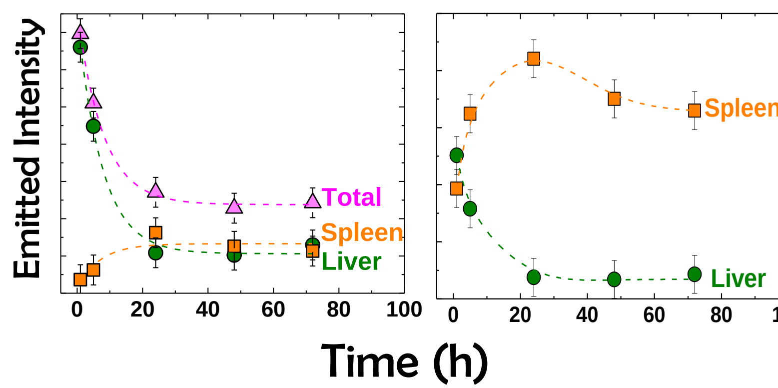
In vivo imaging in the II-BW:





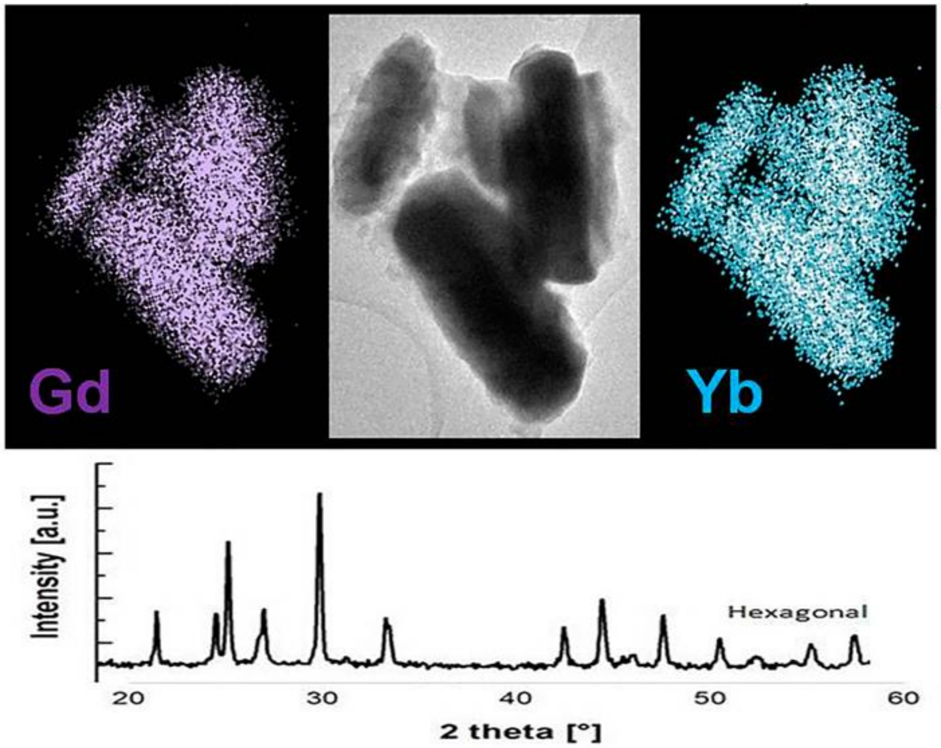
1. IR imaging con NPs signal coming from the abdominal region of the mouse.
2. Only Nd^{3+} luminescence at 1330 nm is not affected by the food light output.
3. Nd^{3+}: SrF_2 NPs enter mouse venous blood reaching organs. Most of the NPs for short time can be found in the mouse liver. Luminescence seems to be rapidly accumulated in the liver and slowly in the spleen. It seems that the luminescence in the liver reduces while the one in the spleen increases with time



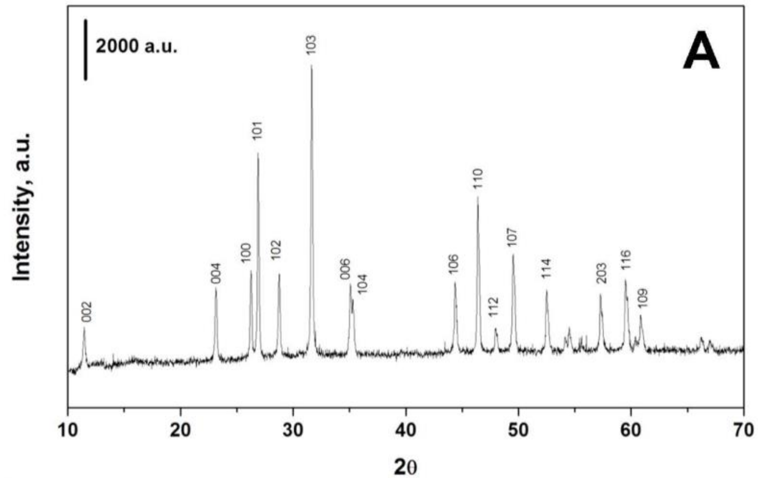


1. The first mouse biodistribution studies of these nanoparticles in mice have been successfully performed.
2. Residual food molecules show a persistent emission in the II-BW, which must be carefully taken into account to perform an accurate imaging. For in vivo mice imaging the problem of fluorescence food is overcome by means of the 1330 nm Nd^{3+} ion emission ($4F_{3/2} \rightarrow 4I_{13/2}$).
3. The results here obtained indicate that SrF_2:Nd nanoparticles are particularly promising for deep tissue fluorescence bioimaging.

Persistent luminescence (PeL) in the near infrared region for medical imaging



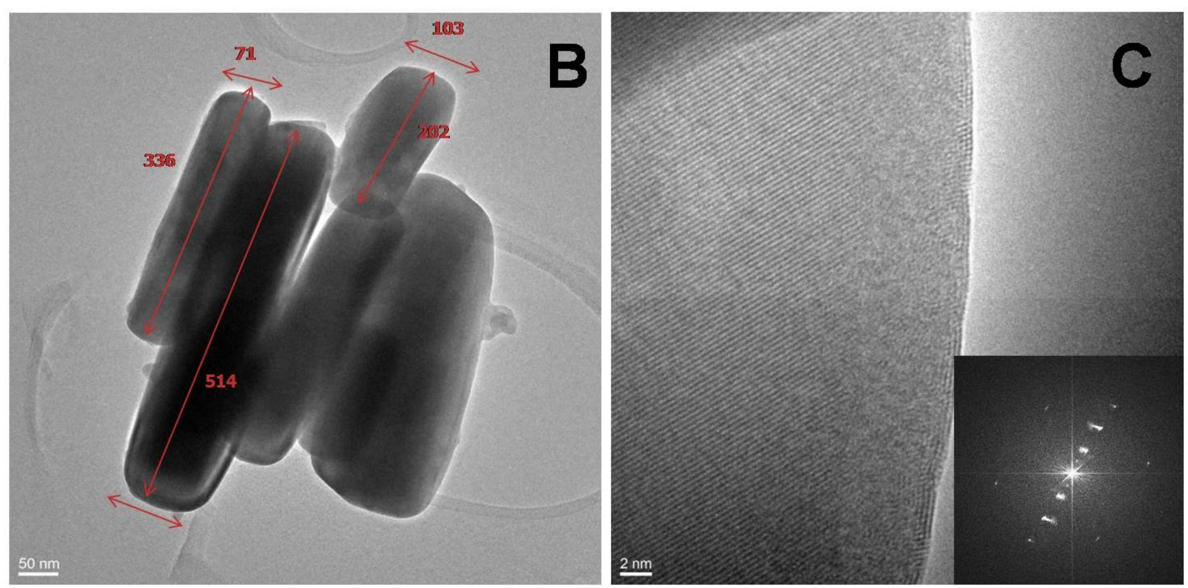
Oxycarbonate ($\mathbf{Gd_2O_2CO_3}$:RE)



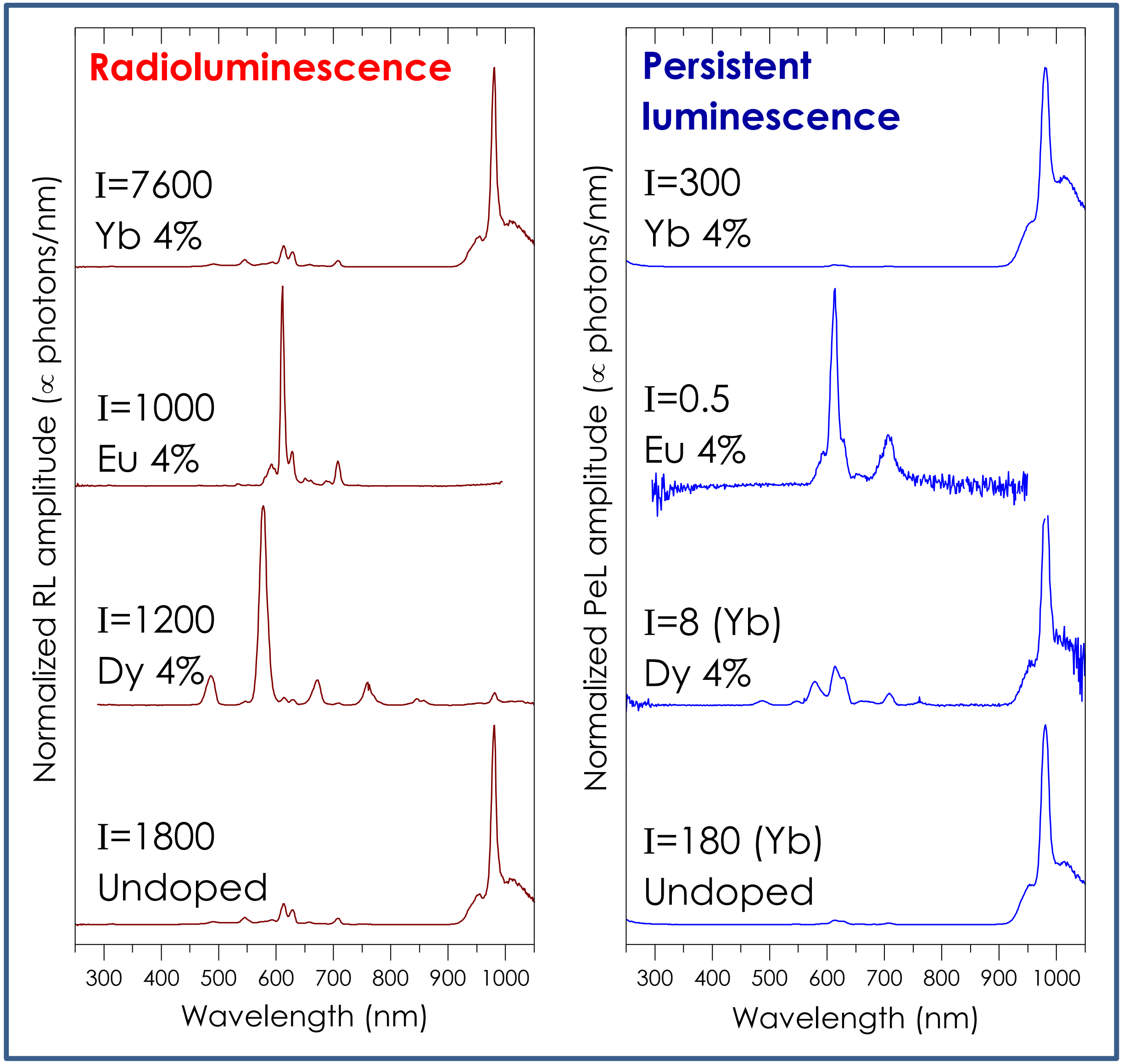
$Gd_2O_2CO_3:Yb^{3+}$

- RE*-doped Gd oxycarbonates where prepared by the Genova University using the hydrothermal synthesis
- Formation of hexagonal phase with high grade of crystallinity.
- Sub-micrometrical particles with rod-like morphology.

*RE = Yb^{3+}, Eu^{3+}, Dy^{3+}, Ho^{3+}



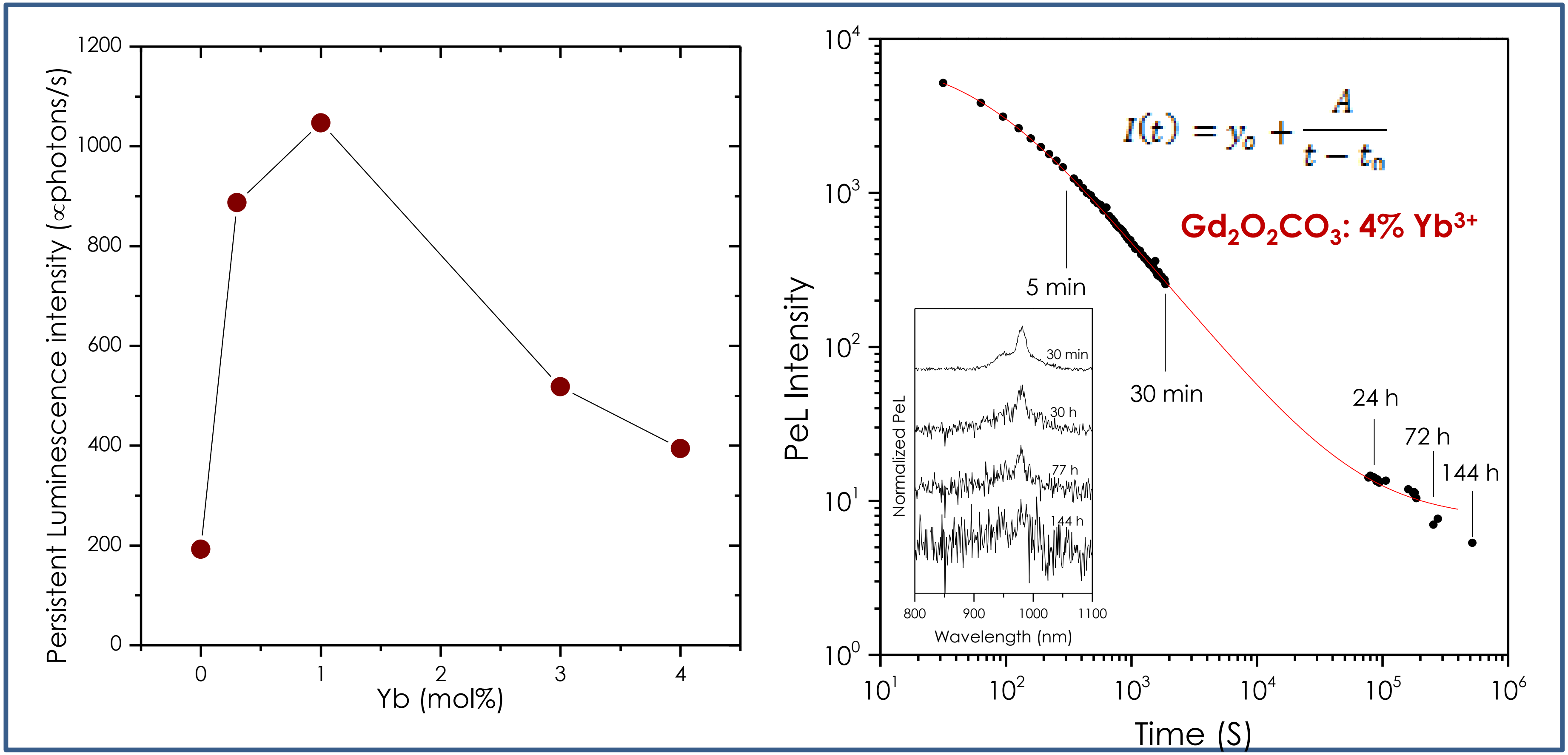
$Gd_2O_2CO_3$:RE RL vs PeL



- RL spectra show the characteristic emissions of the dopant ion.
- Only Yb^{3+} emission was found to produce a PeL emission.
- For the Eu^{3+} doped sample only a short lived afterglow was detected .

$Gd_2O_2CO_3$:Yb^{3+} PeL

- The highest PeL emission was detected for the Yb 1mol% doped sample (**lower** than the one from **$SrAl_2O_4$:Eu^{2+} Dy^{3+} by a factor 40**).
- For **Yb 4 mol%** doped sample The PeL was detectable after up to **140 h (6 days)**.



- Strong luminescence signal in the infrared spectral region.
- Optimal Yb concentration of 1 mol%.
- Persistent signal following x-ray excitation (more than 3 days).
- Potential application of **$Gd_2O_2CO_3$**:Yb^{3+} for *in vivo* imaging

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1.3 μm emitting $SrF_2:Nd^{3+}$ nanoparticles for high contrast *in vivo* imaging in the second biological window

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NIR Persistent Luminescence of Lanthanide Ion-Doped Rare-Earth Oxycarbonates: The Effect of Dopants

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