

Nanoparticles for Biomedical Application

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Core-shell nanostructures have a great potential for biomedical applications.

The goal of this work was to produce these nanostructures from commercial polystyrene nanospheres, used as core, and recover them with a silica shell. Several parameters were analyzed to optimize the experimental procedure, being the superficial charge of the polystyrene nanospheres one of the main parameters.

Different treatments were used to remove the polystyrene core and produce hollow silica nanospheres. These nanospheres were doped with erbium fluoride (ErF_3) and ytterbium fluoride (YbF_3) to analyze the up conversion emissions through photoluminescence characterization. With this analysis it was observed that the sample with 1 % mol. of ErF_3 and 7,5 % mol. Of YbF_3 was the one that had a more intense up conversion emission. With the best co-doped sample it was also analyzed the energy back transfer between erbium and ytterbium and the response to different power inputs.

We hope that this work will be useful for the development of nanoparticles for biomedical application, specifically for bio-imaging area.

KEYWORDS: Nanospheres; Nanostructures; Sol-Gel; Up conversion; Photoluminescence; Bio-imaging.

The presence of nanotechnologies in biomedical sciences provides the development of nanostructures to improve the diagnostic and therapy of several diseases. One of these examples is the use of nanoparticles for bio-imaging [1-3].

The up conversion effect is an alternative process to convert infrared radiation into visible radiation and is based on the sequential absorption of photons [4]. Nanoparticles where this effect is present are very promising for different applications in bio-imaging.

In this work, silica nanoparticles were used because they are chemically inert and, doping with rare-earth ions, is possible to promote the up conversion effect. To prepare the silica nanoparticles in colloidal systems the sol-gel method was used [5].

The first step of this work was the preparation of core-shell nanospheres, with a core of polystyrene and a silica shell. We started with a dispersion of commercial polystyrene

nanospheres, with 460 nm of diameter, and used the experimental procedure described on the Figure 1, according to D'Acunzi et al [6].

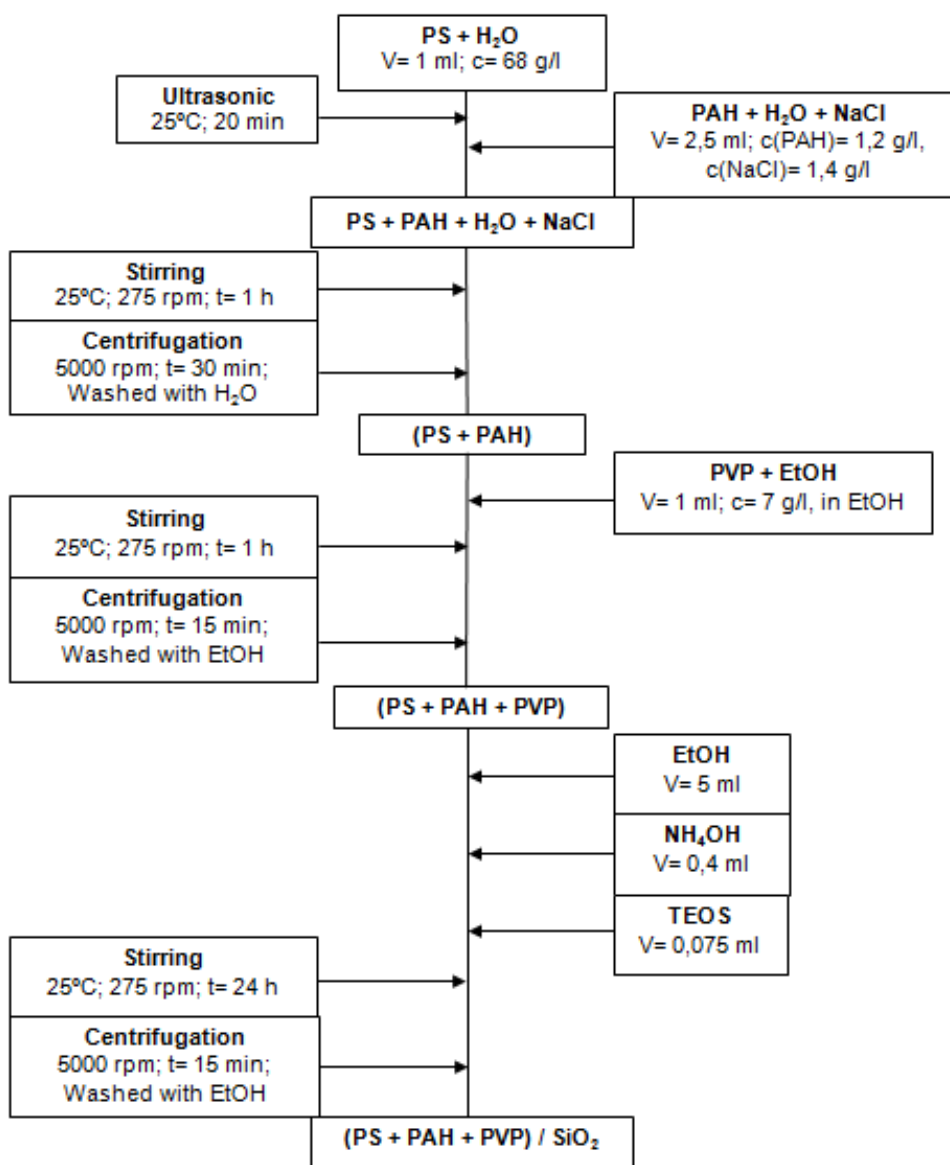


Figure 1 – Experimental procedure used to produce the core-shell nanospheres, with a core of polystyrene and a silica shell, according to D'Acunzi et. al [6].

In this procedure poly (allylamine hydrochloride) (PAH) was added to change the superficial charge of the polystyrene nanospheres, from anionic to cationic, in order to achieve a uniform adhesion of silica, that as a negative charge. Polyvinylpyrrolidone (PVP) was used as a stabilizer to promote the growth of the silica shell on the surface of the polystyrene nanospheres. Ammonium hydroxide (NH₄OH) was used as a catalyst and tetraethylorthosilicate (TEOS) as silica precursor.

The core-shell nanospheres were observed by Transmission Electron Microscopy (TEM). Figure 2 shows the nanospheres with a silica shell thickness between 50 and 60 nm.

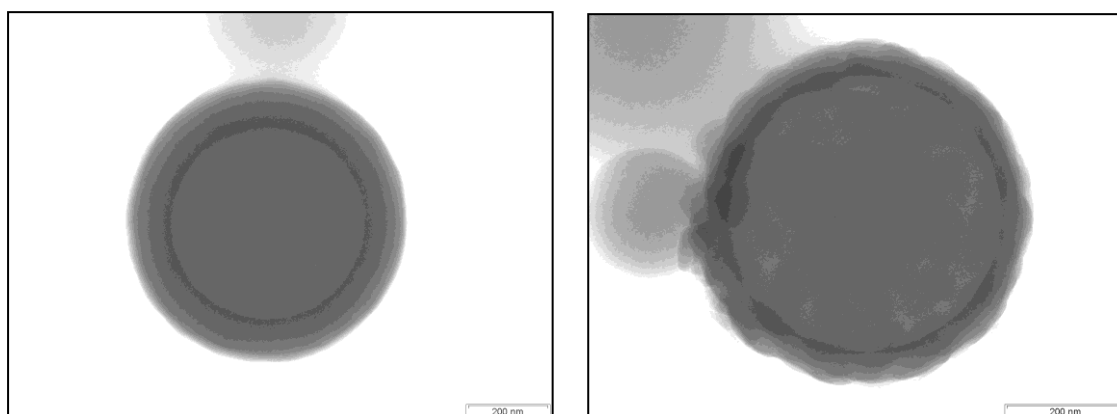


Figure 2 – TEM images of the obtained core-shell nanospheres.

The second step of this work was to produce hollow silica nanospheres, by removing the polystyrene core. To achieve that we used two different treatments: dissolution with cyclohexane and calcination at 450 °C. Figure 3 shows the hollow silica nanospheres obtained after these two treatments.

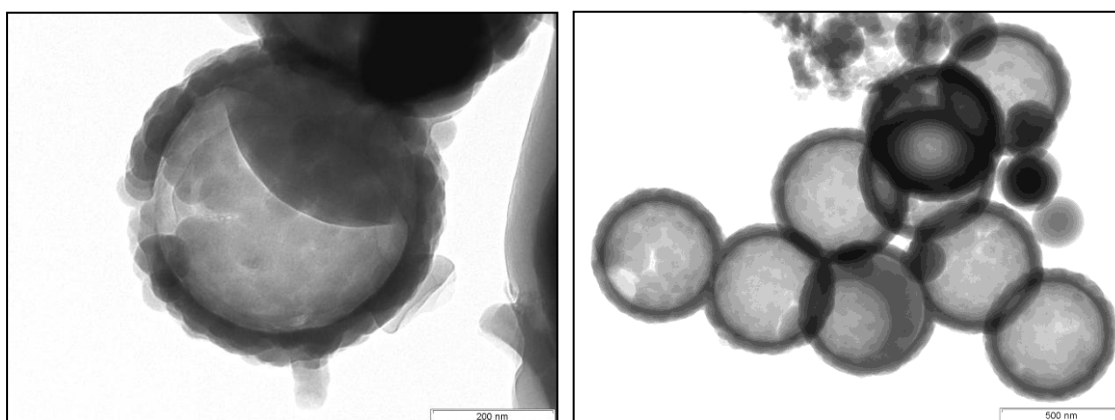


Figure 3 – TEM images of the hollow silica nanospheres. On the left obtained by dissolution with cyclohexane and on the right obtained after a calcination at 450 °C.

To determine the efficiency of each treatment four samples were analysed by Fourier Transform Infrared Spectroscopy (FTIR): i) commercial polystyrene nanospheres; ii) core-shell polystyrene-silica nanospheres; iii) hollow silica nanospheres obtained by dissolution with cyclohexane; iv) hollow silica nanospheres obtained by calcination at 450° C. In Figure 4, the transmission spectra of the samples are shown.

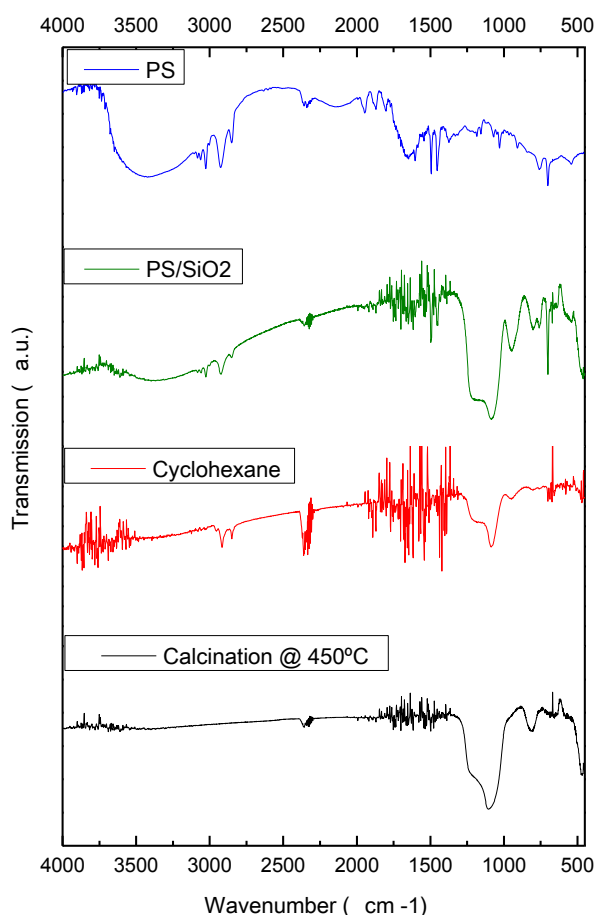


Figure 4 – FTIR spectra of the commercial polystyrene nanospheres, the core-shell polystyrene-silica nanospheres, the hollow silica nanospheres obtained by dissolution with cyclohexane and the hollow silica nanospheres obtained after a calcination at 450 °C.

The characteristic bands of the polystyrene nanospheres are clearly seen at 3000, 1470 and 765 cm^{-1} . These bands are present in the polystyrene nanospheres and in the core-shell polystyrene-silica nanospheres. Analyzing the two treatments we can see that the hollow silica nanospheres obtained by dissolution with cyclohexane still have a residual amount of polystyrene. On the other hand the hollow silica nanospheres obtained after a calcination at 450 °C left no trace of polystyrene at all. So we can conclude that the calcination is more efficient to remove the polystyrene core and produce the hollow silica nanospheres, what is in agreement with the literature [7].

In the final step of this work hollow silica nanospheres, doped with erbium fluoride (ErF_3) and ytterbium fluoride (YbF_3), were produced. The rare-earth ions were introduced 30 minutes after the addition of TEOS. To remove the polystyrene core a calcination at 900 °C was made. At this temperature the OH groups were eliminated, avoiding the presence of humidity and organic groups that could reduce the efficiency of the photoluminescence characterization.

It's usual to co-doped materials with erbium and ytterbium to improve the erbium efficiency in terms of light absorption. This happens because the ytterbium absorbs very efficiently the infrared radiation and then transfer the energy to the erbium ions improving the erbium emissions in the visible, and then we can obtained a much more intense up conversion effect [8].

Finally, in this work, the co-doped hollow silica nanospheres were characterized by photoluminescence. To analyze the up conversion effect five samples were prepared to optimize the amounts of rare-earth ions in order to compare the different intensities of the up conversion effect. Table 1 shows the different amounts of ErF_3 and YbF_3 used in the five samples.

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Name	Er1Yb5	Er2Yb5	Er0,5Yb5	Er1Yb10	Er1Yb7,5
ErF3 (molar %)	1	2	0,5	1	1
YbF3 (molar %)	5	5	5	10	7,5

Table 1 – Different amounts of ErF_3 and YbF_3 used to produce co-doped hollow silica nanospheres.

The entire five samples were excited with a 980 nm wavelength radiation. The obtained spectrum for each sample is shown in Figures 5 and 6.

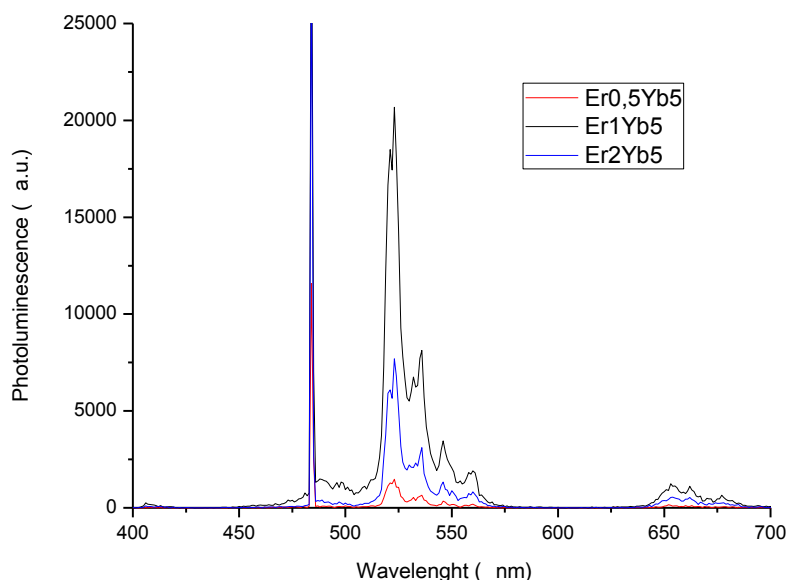


Figure 5 – Photoluminescence spectra, with a source of 980 nm wavelength, of three co-doped hollow silica nanospheres samples: Er0,5Yb5, Er1Yb5 and Er2Yb5.

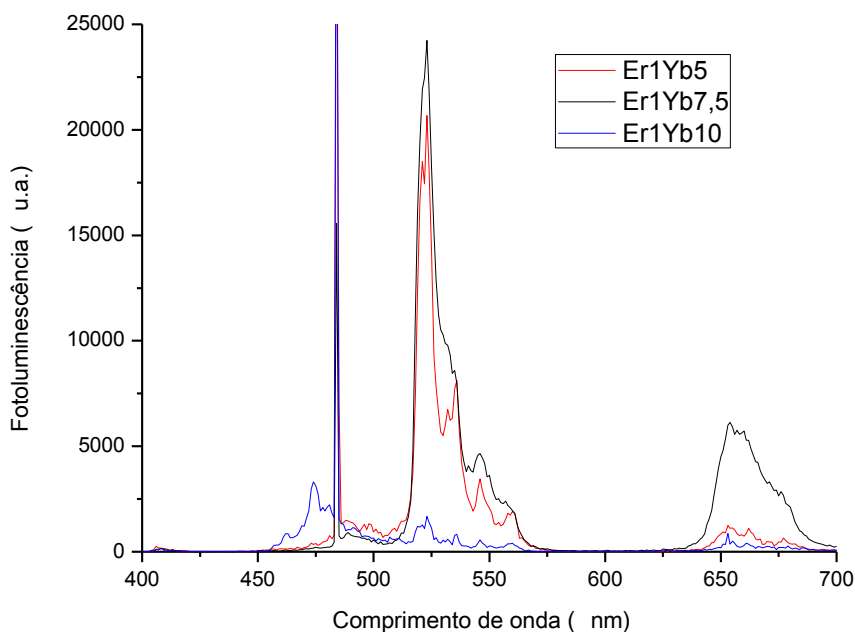


Figure 6 – Photoluminescence spectra, with a source of 980 nm wavelength, of three co-doped hollow silica nanospheres samples: Er1Yb5, Er1Yb7,5 and Er1Yb10.

With the analysis of the obtained spectrum it's possible to see the three erbium emissions in the visible: blue (~484 nm), green (~523 nm) and red (~654 nm). Comparing all the five samples we conclude that the sample Er1Yb7,5 was the one that showed a more intense up conversion effect. So, with this sample, the response to different power inputs was analyzed in order to understand the number of photons involved in the energy transfer between ytterbium and erbium for the three different visible emissions: blue, green and red. For this attempt we used different power inputs, laying from 0,44 to 4 W, and collected the different intensities of the sample. With the obtained points a linear fit was made and the slope of the different graphics was determined in order to calculate the number of photons involved in each visible emission. Figure 7 shows the three graphics obtained for each emission.

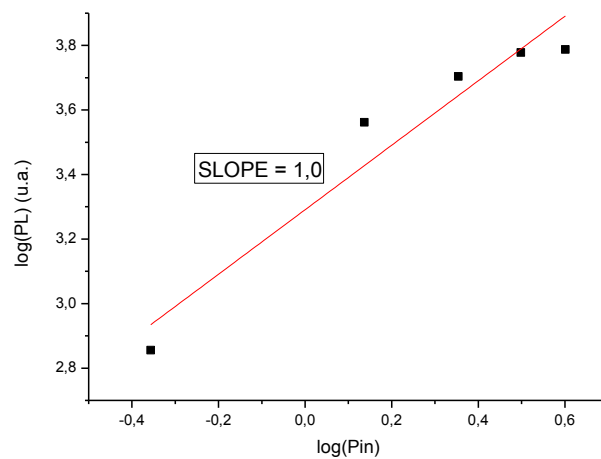
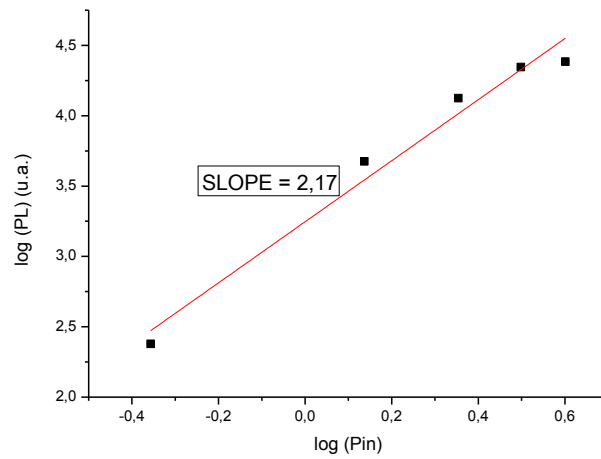
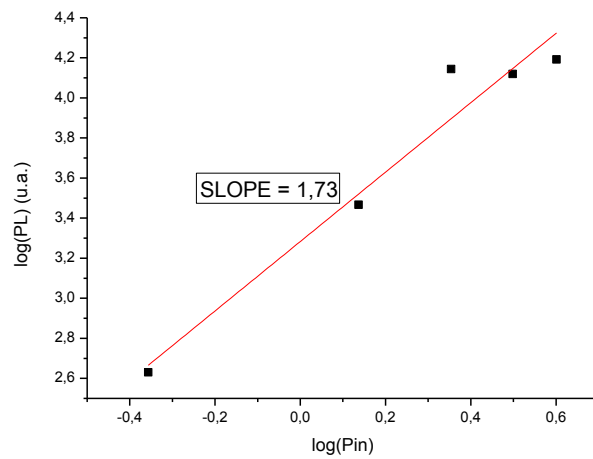


Figure 7 – Graphics obtained with a source of 980 nm wavelength, at different power inputs. Starting from the top, the first one corresponds to the blue emission, the second to the green emission, and the third to the red emission.

Analyzing the three graphics it's possible to conclude that, in the blue and green emission, two photons were involved in the energy transfer process between ytterbium and erbium. For the red emission just one photon was involved in the process. This value is lower than the others maybe because the needed energy to transfer from ytterbium to erbium is not so high. To verify this fact we need to make another test with more different inputs in order to obtain more points to determine a reliable value for the slope.

With the Er1Yb7,5 sample another characterization was made to see the efficiency of the energy transfer between ytterbium and erbium. This characterization was related with the energy back transfer between the two rare-earth ions. For that we used a laser with a wavelength of 514 nm in order to analyze the erbium emission at 1550 nm. In Figure 8 is shown the resulting spectrum of this analysis.

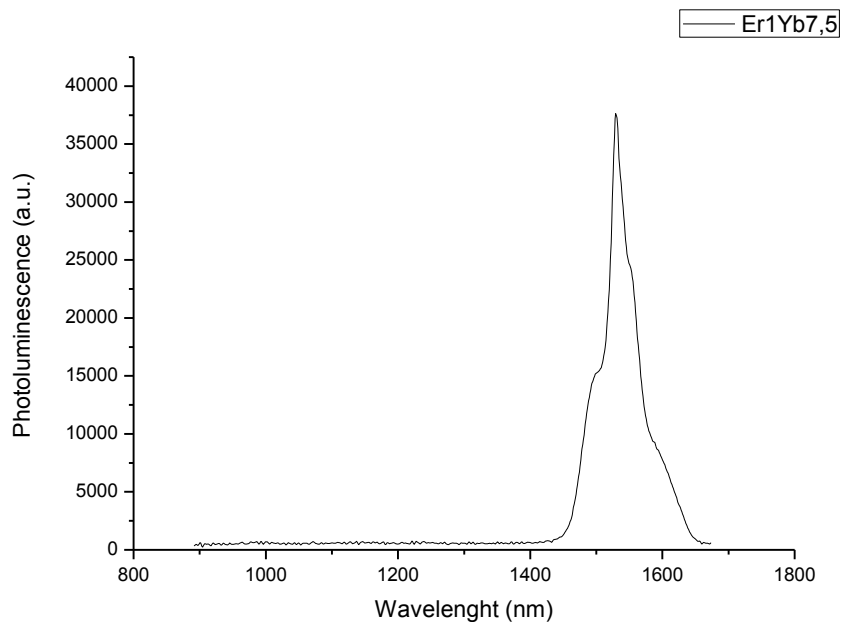


Figure 8 – Photoluminescence spectra of the Er1Yb7,5 sample, with a source of 514 nm wavelength, using an input power of 800 mW.

With this spectrum we can observe the erbium emission at 1550 nm and the ytterbium emission at 1030 nm is not observed. This fact could indicate that all the radiation was absorbed by the erbium ion and so the energy back transfer didn't take place, which indicates that the efficiency of the up conversion effect was high. But, to conclude with any doubts that the energy back transfer didn't occur, we need to analyze monodoped samples, ones just with erbium, and others just with ytterbium.

The first conclusion of this work is the fact that the superficial charge of the polystyrene nanospheres strongly affects the silica coating. To obtain core-shell nanospheres the addition of PAH is needed to change the superficial charge of the nanospheres, from anionic to cationic, in order to coat the spheres with silica, that as an anionic charge.

Another conclusion involves the production of hollow silica nanospheres. To remove the polystyrene cores the most effective treatment was the calcination at 450 °C, because with this heat treatment all the polystyrene were removed from the nanospheres.

For the co-doped hollow silica nanospheres we concluded that the sample that had a more intense up conversion effect was the sample with 1 % mol. of ErF_3 and 7,5 % mol. of YbF_3 . With this sample we made two different analyses, one with different power inputs in order to know the number of photons involved in the three visible emissions: blue, green and red, and another with an excitation at 514 nm to see the energy back transfer process between the two rare-earth ions. With different power inputs we concluded that for the blue and green emissions two photons were involved in the energy transfer from ytterbium to erbium and, on the other hand, just one photon was involved in the red emission. With the excitation at 514 nm we concluded that the energy back transfer didn't take place and so the efficiency of the up conversion effect was high.

Finally, with all the photoluminescence characterization it's possible to conclude that the core-shell nanospheres produced in this work are capable to use in bio-imaging processes.

References:

- [1] Paras N. Prasad, *Fundamentals of Nanobiophotonics*. Photon-based Nanoscience and Nanobiotechnology, 2006. **239**: p. 55-65.
- [2] Haglund, E.; Seale-Goldsmith, M.M., Leary, J.F., *Design of Multifunctional Nanomedical Systems*. Annals of Biomedical Engineering, 2009. **37**(10): p. 2048-2063.
- [3] Jiang S.; Zhang, Y.; Lim, K.M.; Sim, E.K.W.; Ye, L., *NIR-to-visible upconversion nanoparticles for fluorescent labeling and targeted delivery of siRNA*. Nanotechnology, 2009. **20**(15): p. 155101.
- [4] Liu, G.; Jacquier, B, *Spectroscopic Properties of Rare Earths in Optical Materials*. 2005, Springer.
- [5] Brinker, C.J.; Scherer, G.W., *Sol-Gel Science: The Physics and Chemistry of Sol-Gel Processing*. 1989, Academic Press.
- [6] Zhang, L.; D'Acunzi, M.; Kappl, M.; Auernhammer, G.; Vollmer, D., *Hollow Silica Spheres: Synthesis and Mechanical Properties*. Langmuir, 2009. **25**(5): p. 2711-2717.

[7] Imhof, A., *Preparation and Characterization of Titania-Coated Polystyrene Spheres and Hollow Titania Shells*. Langmuir, 2001. **17**(12): p. 3579-3585.

[8] Ronda, C., *Luminescence: From Theory to Applications*. 2008, Wiley.