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Author
Badaracco, Adrian Garcia

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Low Efficiency Upconversion Nanoparticles for High-Resolution Coalignment of Near-Infrared and Visible Light Paths on a Light Microscope

Adrian Garcia Badaracco

Sriramkumar Sundaramoorthy,† Adrian Garcia Badaracco,‡ Sophia M. Hirsch,§ Jun Hong Park,‡ Tim Davies,† Julien Dumont,¶ Mimi Shirasu-Hiza,§ Andrew C. Kummel,‡ and Julie C. Canman, †*

†Department of Pathology and Cell Biology, Columbia University, New York, New York 10032, United States
‡Section of Chemical and Materials Science, University of California, San Diego, La Jolla, California 92093, United States
§Department of Genetics and Development, Columbia University, New York, New York 10032, United States
¶Institut Jacques Monod, CNRS, UMR 7592, University Paris Diderot, Sorbonne Paris Cité, F-75205 Paris, France

ABSTRACT: One major technical barrier in working with both NIR and visible light on an optical microscope is obtaining their precise coalignment at the imaging plane position. Current techniques require complex setups and software. Photon upconverting particles (UCPs) can bridge this gap as they are excited by NIR light but emit in the visible range. Here, two different UCPs have been identified, high-efficiency micro540-UCPs and lower efficiency nano545-UCPs are compared, and it is found that the lower efficiency nano-UCPs were superior for precise coalignment of the NIR beam with the visible light path consistent with limited particle-to-particle energy transfer, superlinear power dependence for emission, and much smaller particle size.

Materials and Method

The set-up for one-step light path coalignment using UCPs is shown in *Figure 1*. Imaging was performed on a spinning disc confocal microscope, using a CCD camera for imaging in the visible range and an IR sensitive camera for NIR light.

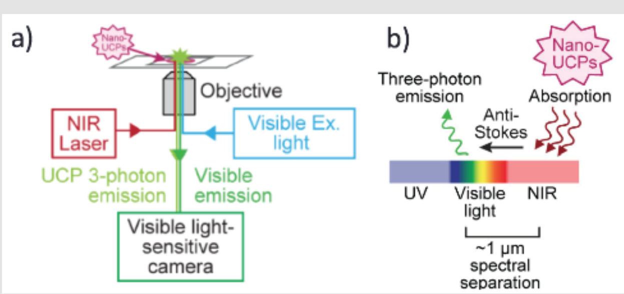

Figure 1. Setup for one step, one camera nano-UCPs for NIR and VIS light path coalignment via UCP emissions (a) and schematic of upconversion process (b).

Material Characterization

Characterization of both types of UCPs is shown in Figure 2. Micro^{540}-UCPs were 2.1 ± 0.07 µm in diameter and EDS analysis showed composition consistent with a rare-earth doped Gd_2O_2S crystal. Nano^{545}-UCPs were on average 27.22 ± 0.81 nm with composition consistent with a rare-earth doped $NaYF_4$ crystal (see *Figure* 2).

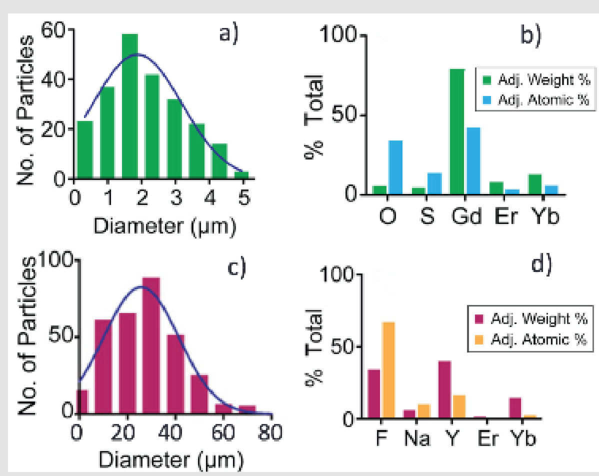

Figure 2. Material characterization of UCP. Size analysis via SEM (a) and EDS analysis of micro540-UCPs. Size analysis via AFM (c) and elemental analysis of nano545-UCPs (d).

Modeling of Power Dep. and Broadening

Power dependence studies were performed on both types of UCPs. Fluorescence intensity at 525 ± 25 nm was quantified following 1470 nm laser excitation and plotted in a double-log plot (see *Figure* 3).

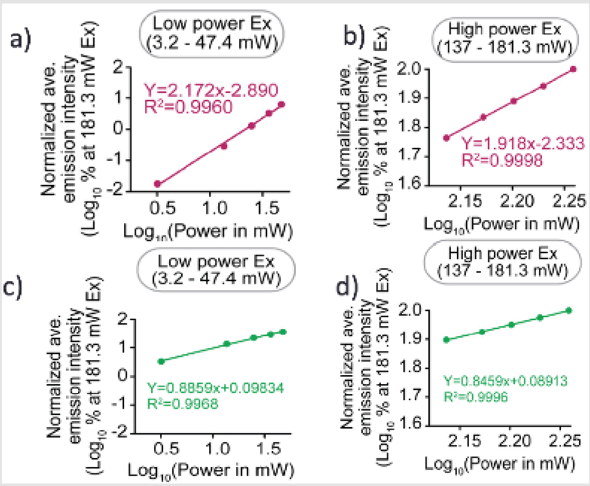

Figure 3. Log-log plots of emission intensity vs. laser power for nano^{538}-UCPs at low laser power (a), high laser power (b) and micro^{540}-UCPs at low laser power (c) and high laser power (d).

For micro^{540}-UCPs, the observed decay in the power dependence from $P^{0.88}$ to $P^{0.85}$ can be attributed to a efficient saturation of the intermediate energy states. In the case of nano^{538}-UCPs, the power dependence is greater with a decay from from $P^{2.17}$ to $P^{1.92}$.

To quantify beam profiles, the laser profile was scaled by the power dependence and then convoluted with a broadening function whose characteristic width is defined as the "effective particle size" (EPS).

This analysis showed that the micro-UCP emission was best fitted using an EPS of 7.78 µm Nano^{545}-UCPs on the other hand showed less beam broadening. These UCPs were best fit by an EPS of 2.33 µm (see *Figure* 4).

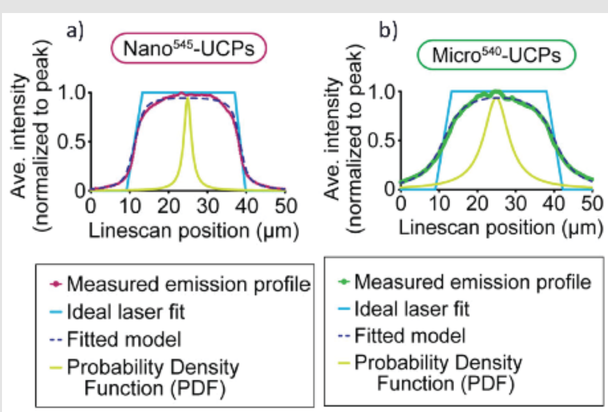

Figure 4. Beam profiles from micro^{540}-UCPs show a broad Lorentzian fit (a) while beam profiles from nano^{545}-UCPs show a tight Lorentzian fit (b).

The effective particle size distribution in both cases takes into account the transfer of excitation between particles (see *Figure* 5); thus, for the purpose of fitting the emission profiles, the particle size distribution is broader than the physical particle size distribution.

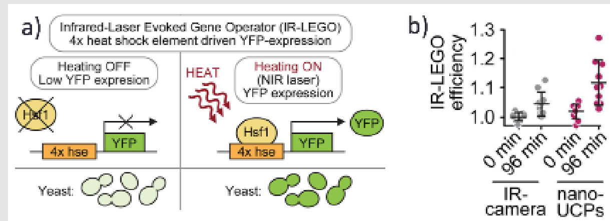

Figure 5. Diagram of practical comparison of coalignment methods using IR-LEGO for YFP expression (a) and IR-LEGO efficiency comparison between coalignment methods (b)

Practical Application

IR heating of yeast cells containing genes for heat shock element driven expression of Yellow FP is performed using either a traditional beam coalignment system or coalignment with nano^{538}-UCPs. Then, relative intensity of emission in the visible yellow channel is used to measure co-alignment. A statistically significant difference between coalignment methods was found ($p = 0.0161$).

Conclusions

UCPs allow a one-step, one camera method to co-align an NIR laser with the specimen plane on a light microscope. Smaller, lower efficiency UCPs were shown to be superior to large more efficient particle based on a model representing an effective optical particle size.

References

Jaba et al. Infrared to Visible up-Conversion Study for Erbium-Doped Zinc Tellurite Glasses. J. Phys.: Condens. Matter 2000, 12 (20), 4523.

Liu, G. Advances in the Theoretical Understanding of Photon Upconversion in Rare-Earth Activated Nanophosphors. Chem. Soc. Rev. 2015, 44 (6), 1635–1652.

Prasad et al. Photopatterned Microstructures Using Lanthanide (Iii) Ion Co-Doped Nayf4 Nanoparticles with Upconversion Luminescence for Possible Applications in Security. Nanotechnology 2009, 20 (18), 185301.

Pollnau et al. Power Dependence of Upconversion Luminescence in Lanthanide and Transition-Metal-Ion Systems. Phys. Rev. B: Condens. Matter Mater. Phys. 2000, 61 (5), 3337.

Sarkar et al. Sub-5 Nm Ln(3)(+)-Doped Baluf(5) Nanocrystals: A Platform to Realize Upconversion Via Inter-particle Energy Transfer (Ipet). Adv. Mater. 2013, 25 (6), 856–60.

Wang et al. Upconversion Nanoparticles in Biological Labeling, Imaging, and Therapy. Analyst 2010, 135 (8), 1839–1854. Achatz, D. E.; Ali, R

Wolfbeis, O. S. Luminescent Chemical Sensing, Biosensing, and Screening Using Upconverting Nanoparticles.Top. Curr. Chem. 2010, 300, 29–50.

Adrian Garcia Badaracco

NanoEngineering

Adrian Garcia Badaracco moved to the US from Argentina in 2012 and quickly developed an interest in engineering & research. He is currently a Senior in Warren college, and will be starting the MS portion of a BS/MS in Chemical Engineering the following school year. He has been working on materials science research under Dr. Kummel at UCSD for over 2 years, and has published in journals such as ACS Applied Materials and Interfaces.

Outside of the lab, Adrian is involved in the Pi Kappa Alpha fraternity, where he leads the education program, and is a member of Tau Beta Pi, foremost engineering honors society. He competes in ice/roller hockey on the school's D2 teams, and plays soccer on the weekends.

Adrian hopes to be able to reuinite with his home country and share some of the valuable knowledge he's earned with his peers.

What has been your greatest challenge in research?
I feel that my greatest challenge is remaining objective and taking and remaining open to new points of view. It is easy to get excited about results or a model and lose track of inconsistencies or issues that could be fundamental to interpreting the data.

What advice would you give to your first-year self?
Work hard and remember that things will not happen overnight.

What excites you about next school year?
I will be taking my last few CENG courses and I will have a couple completely elective (i.e. just for units) courses to take, which should be fun and a good change of pace.

If you were a PI, what research would your lab focus on? And why?
If funding were no problem, I would focus on production of animal products via large scale bioreactors. This is an attractive ideas because it aligns with my family's ideals and I believe it has the potential to be disruptive to the whole food supply industry.

What do you remember from Warren Writing?
I remember having to learn not just how to improve my own work, but also the value in helping others improve theirs, not only in terms of personal fulfillment but also for the benefit and advancement of one's own writing.

How do you keep your life in balance?
I like to exercise regularly and am an active member of several organization on campus. When time allows, I like to undertake small home projects and continually keep myself busy.

Is there anything you wish you could change or do differently from the time you entered college?
I think I am have gotten very lucky and a lot of the choices I made have panned out. I don't really believe in dwelling over past choices: hindsight is always 20-20. If there's one thing I would have liked to tell myself, it's that we had a hockey team (I did not find out until my second year).

What's the coolest research project project you've seen/ heard of, and where did you see it?
I am always excited to hear about Kristine Khieu's (BENG) research in the Hargens lab. As a kid, I wanted to be an astronaut, so it's super cool to hear about all physiological adaptations that the human body undergoes when exposed to conditions in space.