Multispectral optical tweezers for molecular diagnostics of single biological cells

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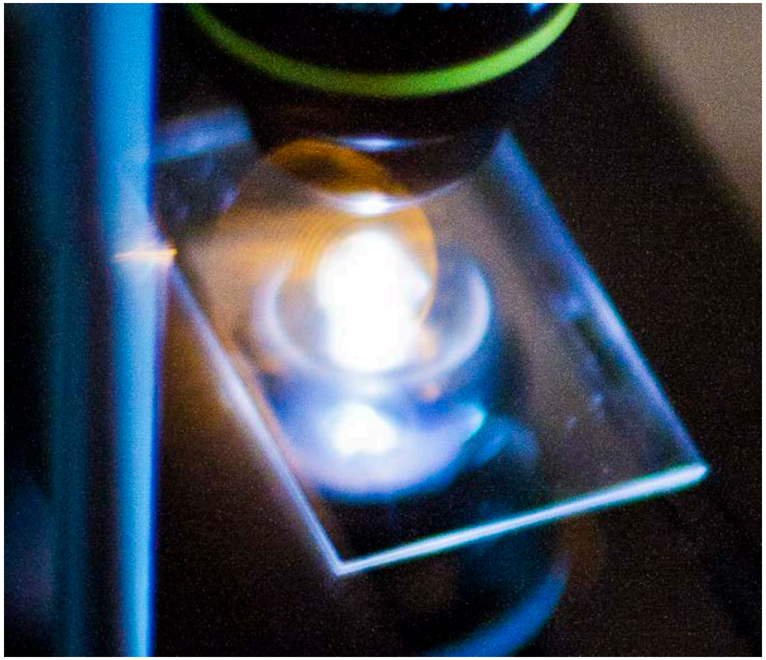
BiOS at Photonics West 2012
Imaging, Manipulation, and Analysis of Biomolecules, Cells, and Tissues X
Saturday, January 21st 2012
San Francisco, CA, USA

Outline

Motivation

Multispectral optical tweezers

Results on yeast cells

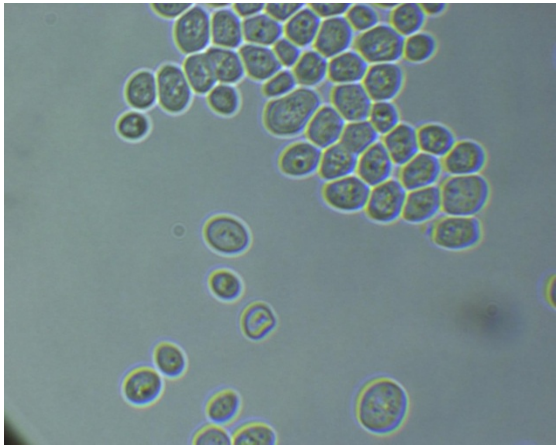


Motivation

Need for single cell manipulation

Avoid (control) many-body effects

- chemical signaling
- interference of diagnostics



Subtract interferences from solvent

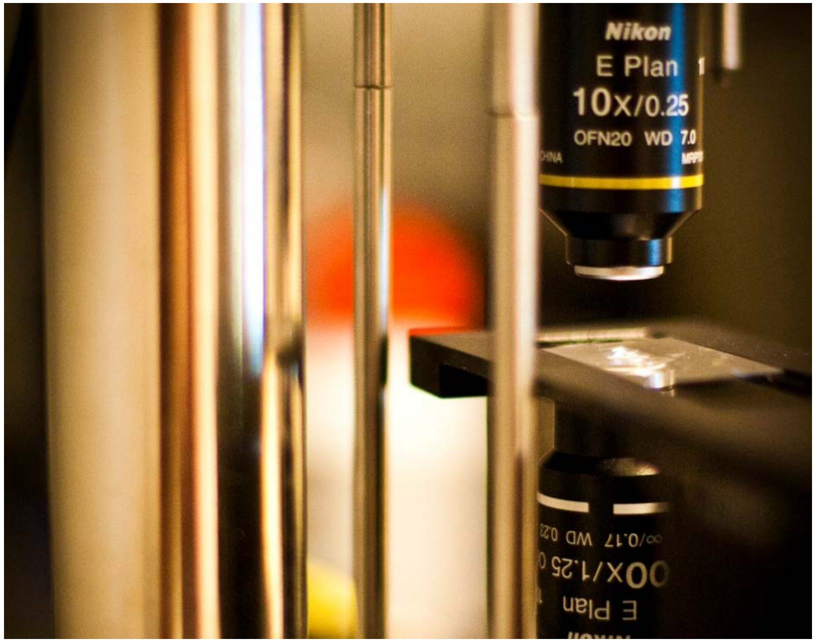
Manipulate a single cell and its environment

Need for full spectral information

Molecular information:

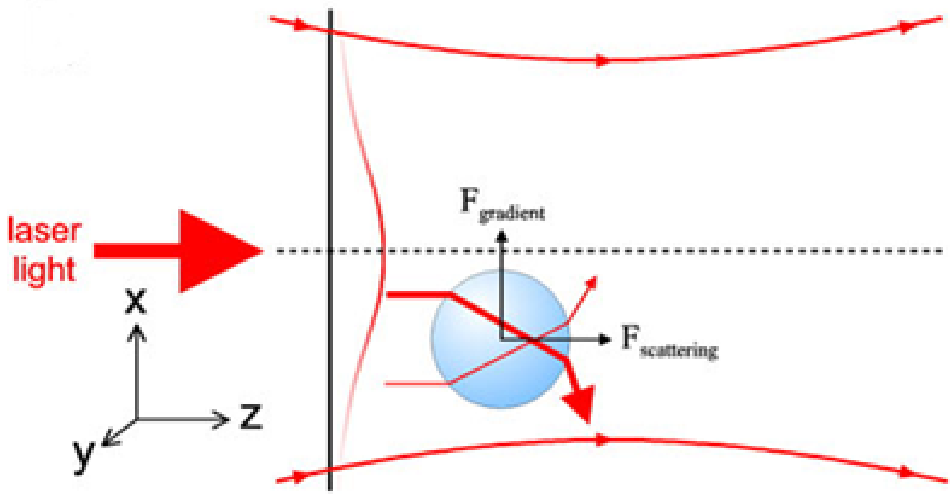
- Electronic: **Fluorescence**
  - High yield but sensitive to environment
- Vibrational: **Raman**, infrared
  - Stability of the signature but low yield

Combination of information from both in a SINGLE product (laser and detector)



Multispectral optical tweezers

Optical trapping



$\text{Ø}_{particle}$ >> λ_{laser} : Mie regime

approx. by ray optics

$\text{Ø}_{particle}$ << λ_{laser} : Rayleigh regime

gradient + scattering forces

$$F_{grad} = \frac{-(n_m r)^3}{2}\left(\frac{m^2-1}{m^2-2}\right)\nabla E^2$$

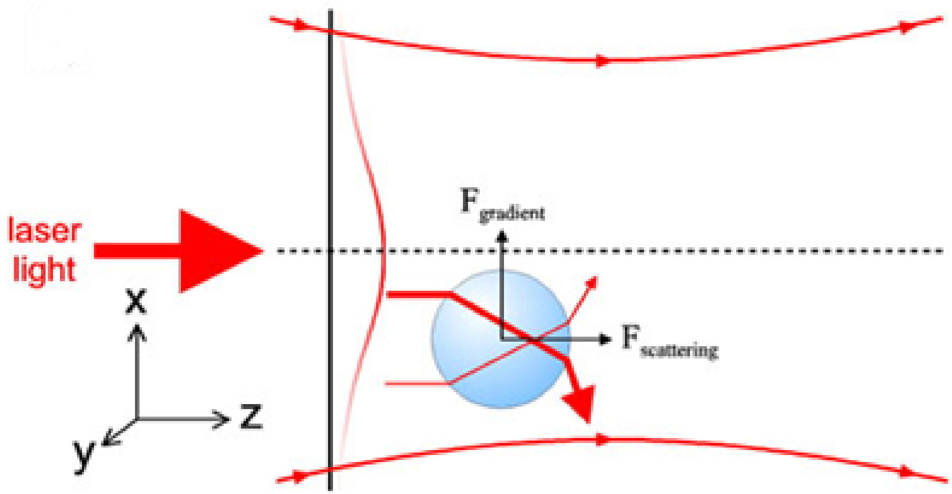
$$F_{scat} = \frac{I_0}{c}\frac{128\pi^5 r^6}{3\lambda^4}\left(\frac{m^2-1}{m^2+2}\right)^2 n_m$$

$m = n_p/n_m$

$n_{Particle} > n_{Medium}$: attraction

$n_{Particle} < n_{Medium}$: repulsion

Optical trapping



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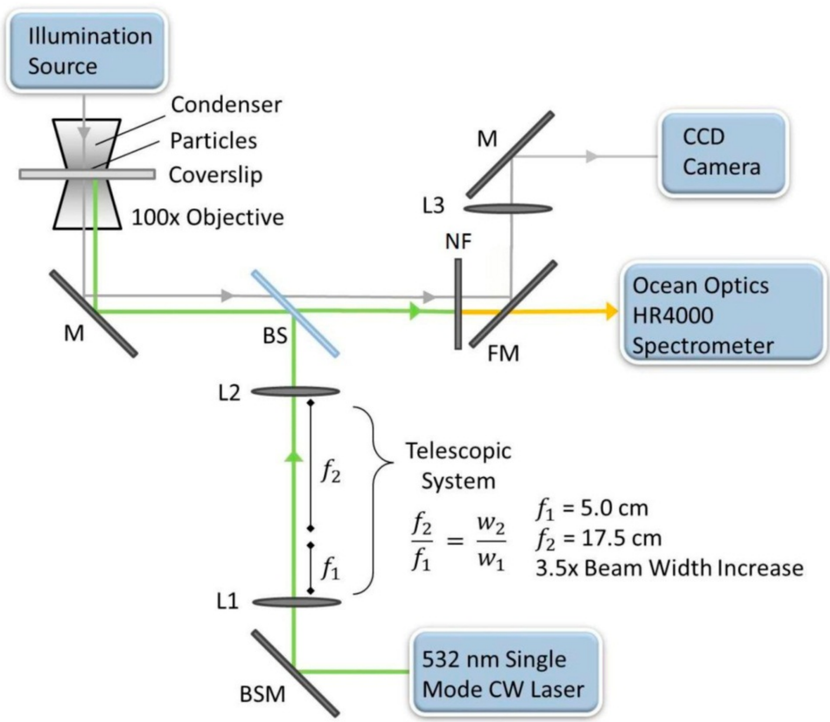
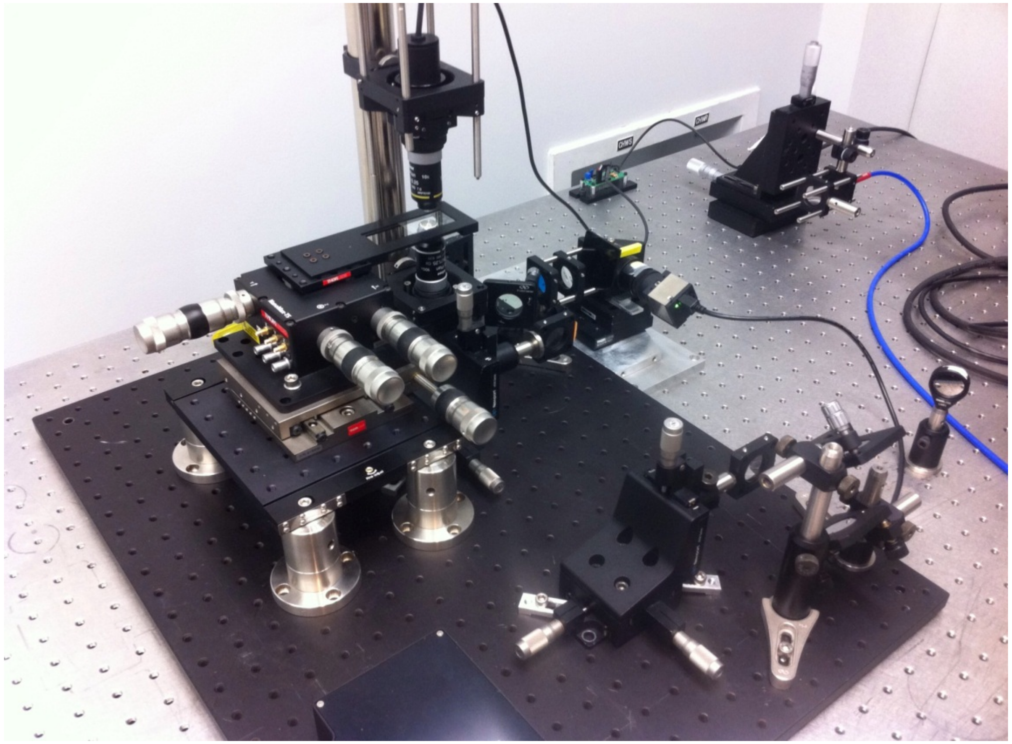
Experimental setup





Based on OTKB (Thorlabs)

Simple multispectral optical tweezers for vibrational AND electronic spectroscopy

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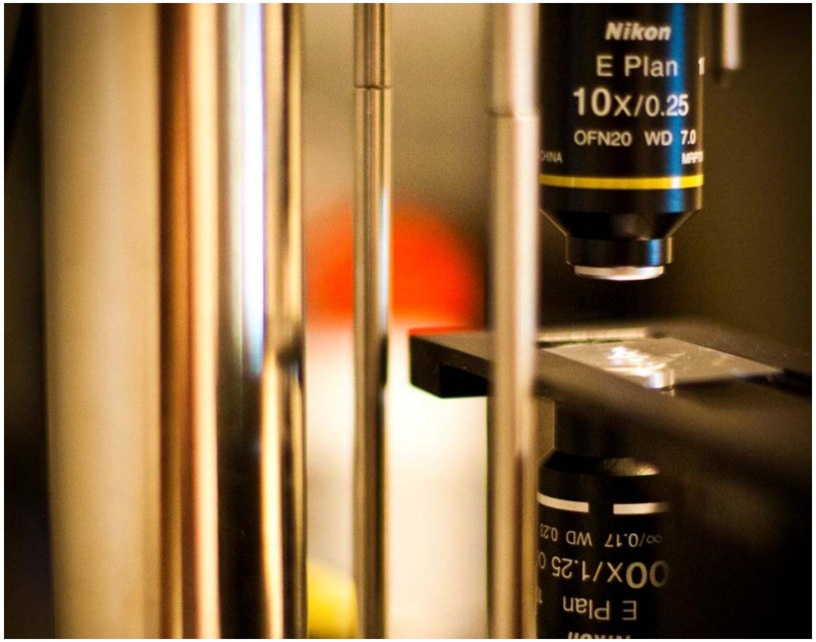
Experimental setup

Laser: Laserglow Aries-20
CW, λ: 532 nm
Power: 5 -30 mW
Single mode

Focusing microscope objective:
100x Oil E Plan Achromat Objective
NA 1.25, WD: 230 μm
Field of View: 25mm

Illumination microscope objective:
10x E Plan Achromat Objective
NA 0.25; W.D. 7mm



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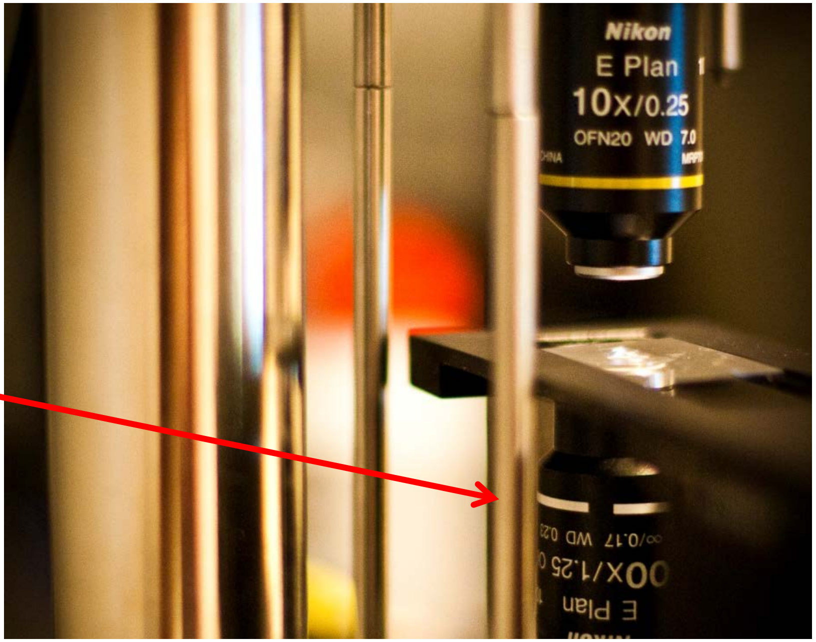

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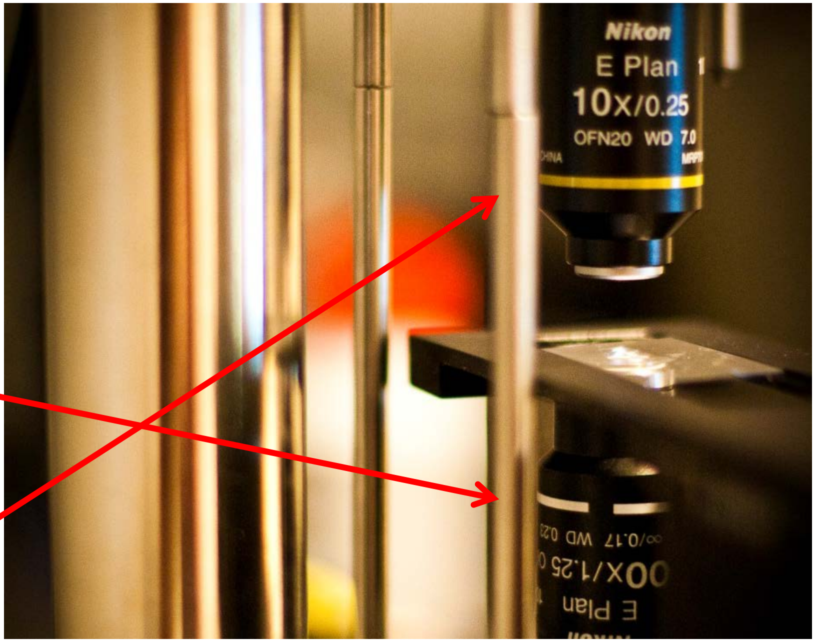

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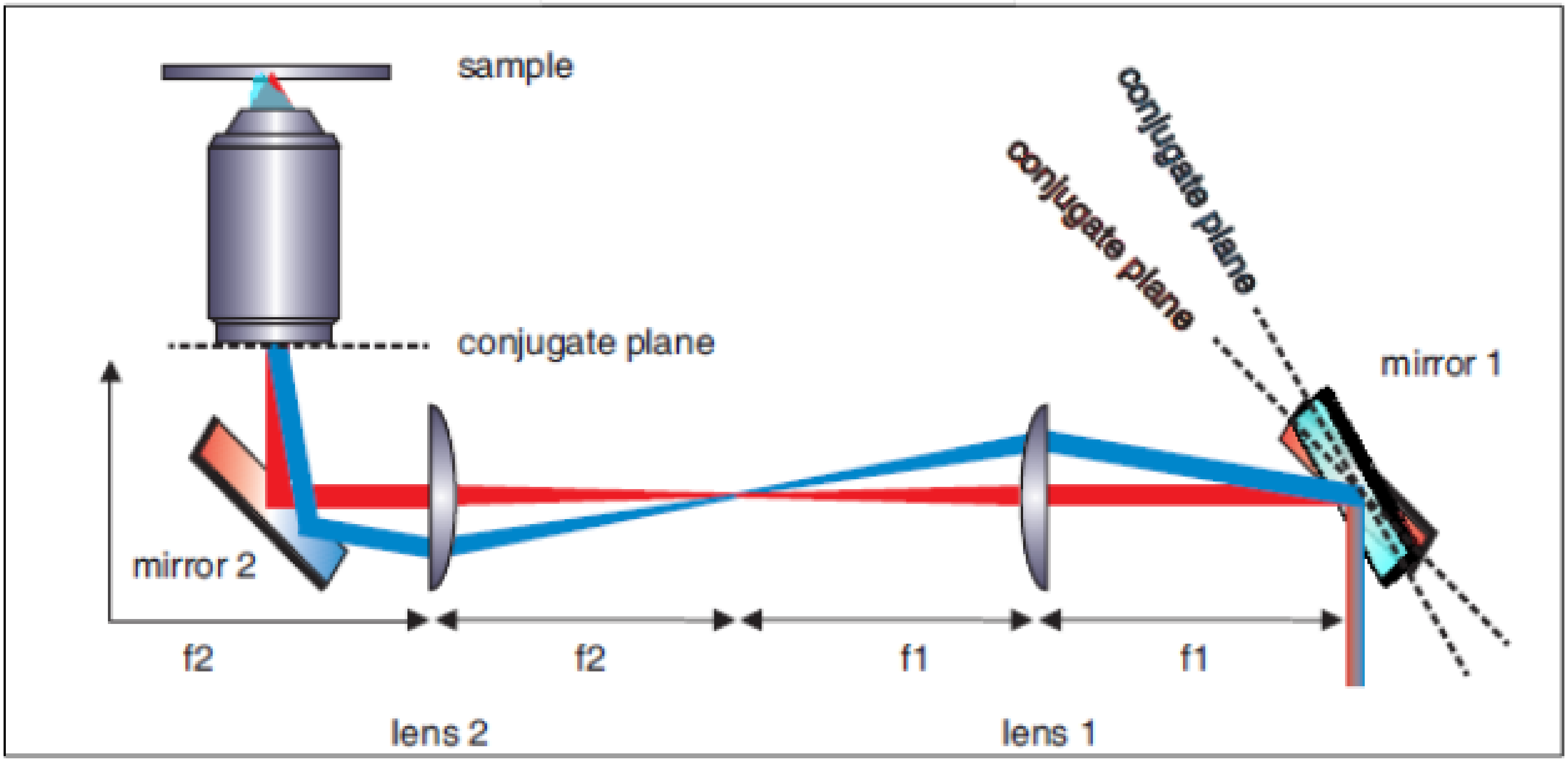
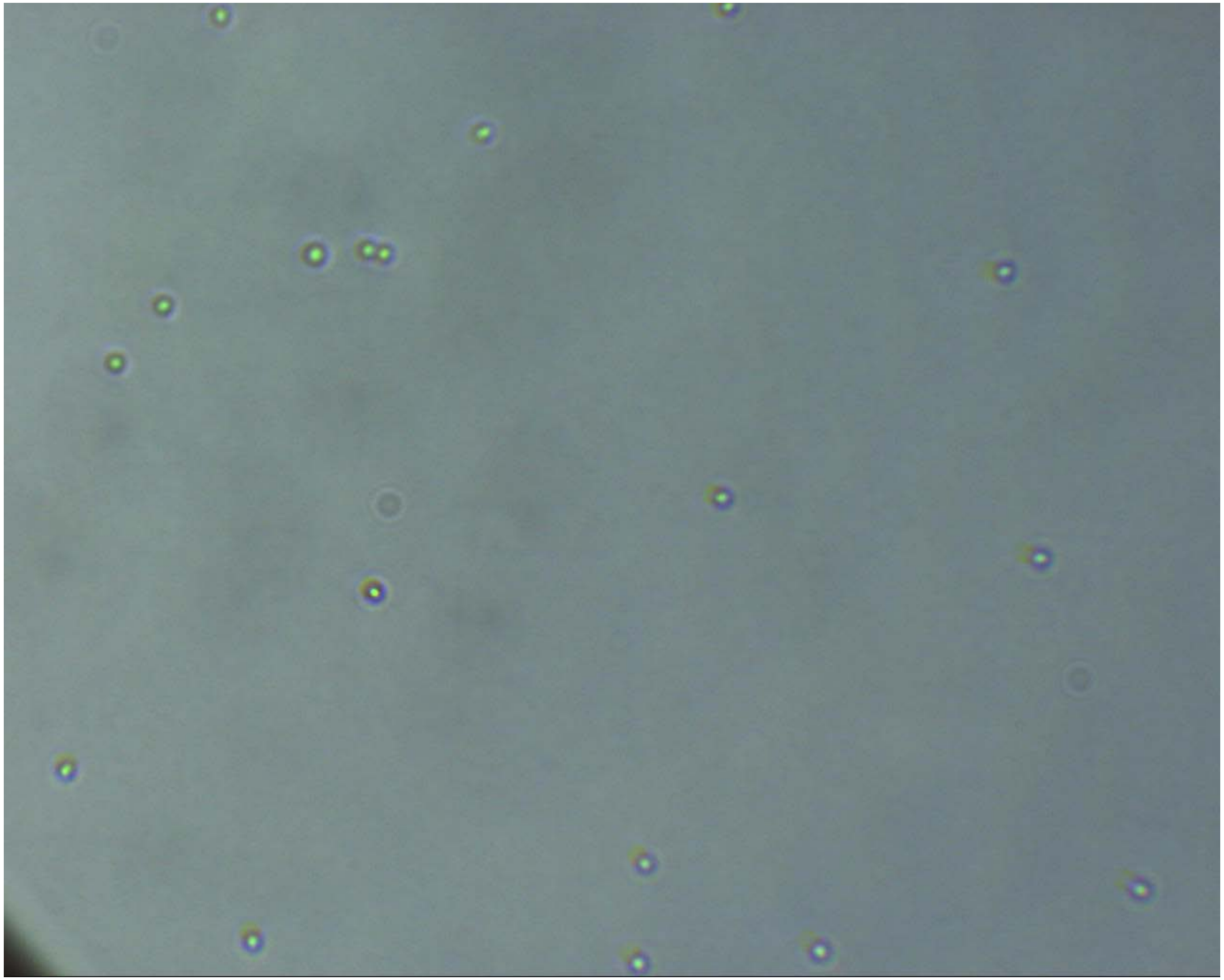
Beam steering



Multiple purpose: - telescope (3.5:1 – f_1=5 cm, f_2=17.5 cm)
- beam steering

Characterization: steering



10 µm

1 µm polystyrene beads

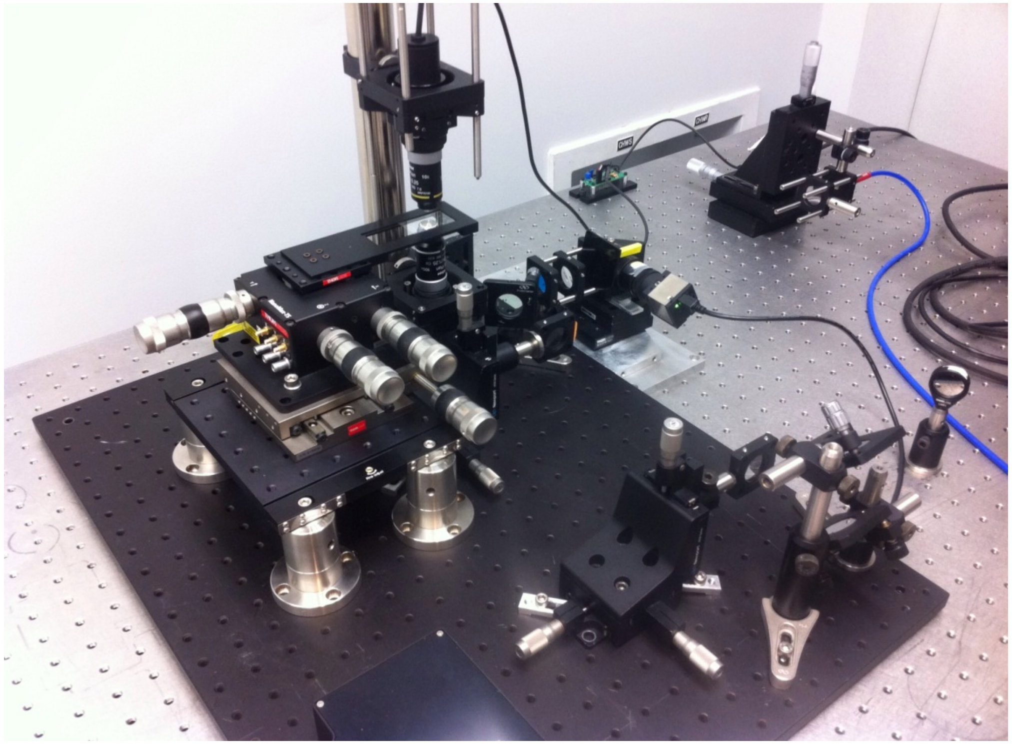
Stable control of the cell/particle over more than 50 µm

Spectral detection

Common spectrometer
Ocean Optics HR 4000

Long-pass optical filter
Semrock LP03-532RS-25

Samples:
Sigma Aldrich LB30-1ML
3µm PS beads (Raman)
Sigma Aldrich L3030-1ML
2µm dyed fluorescent PS beads



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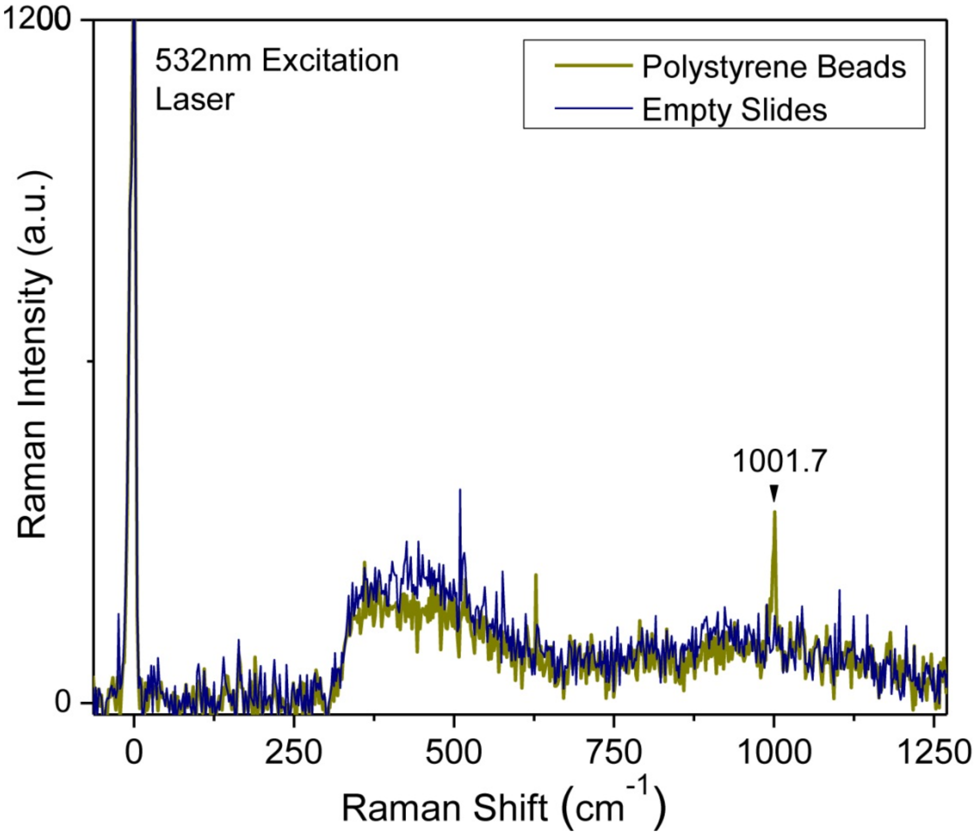
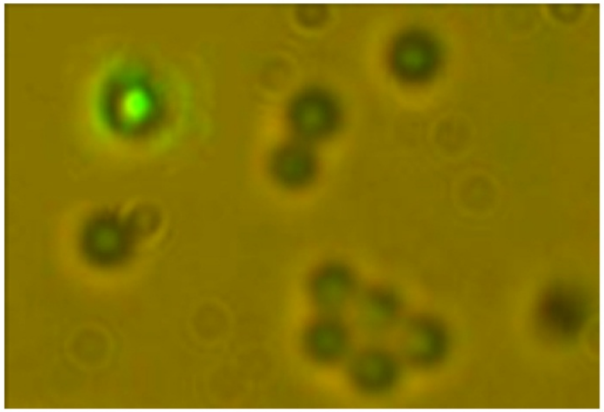
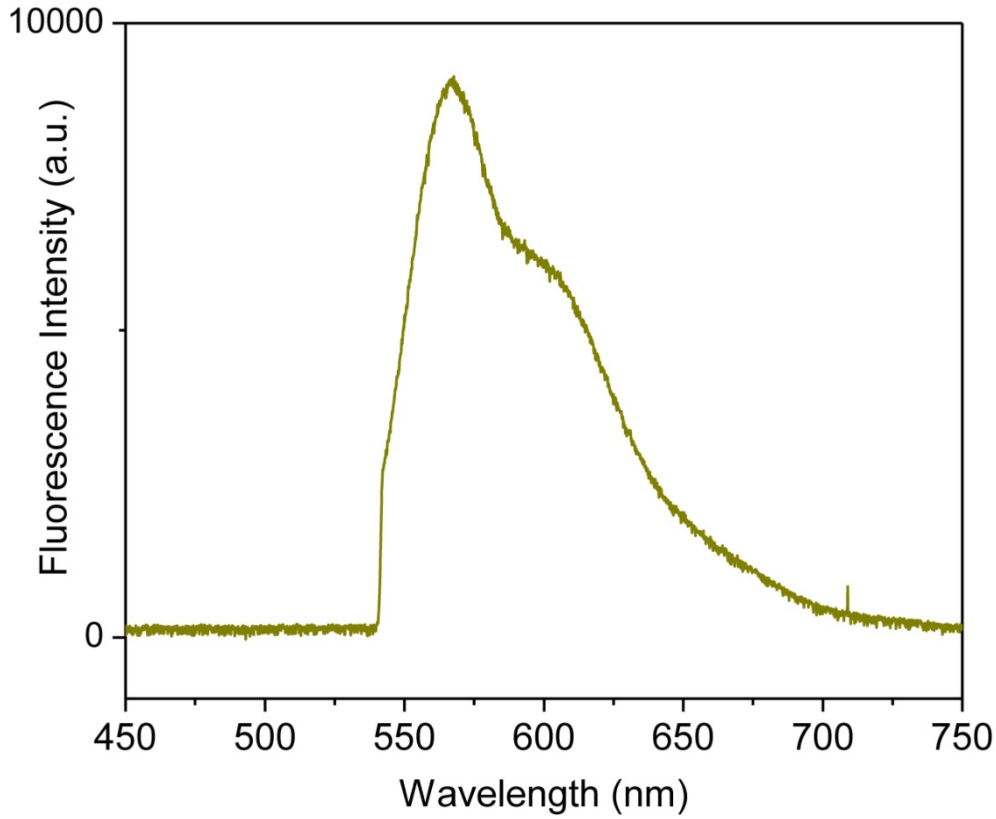
Characterization: Raman

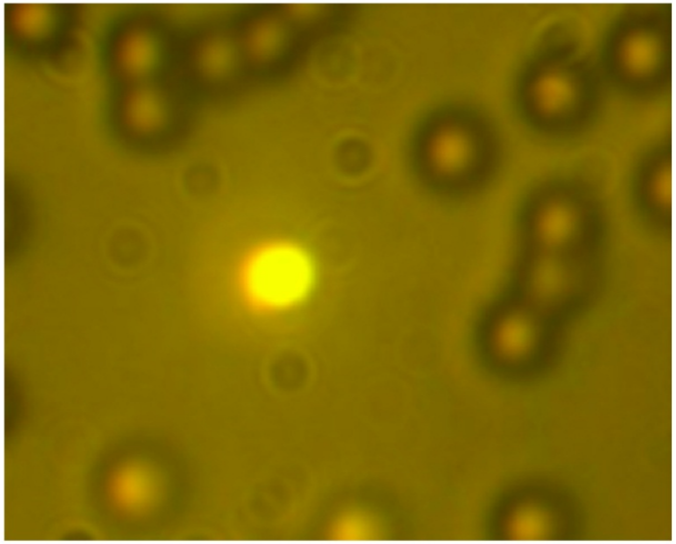




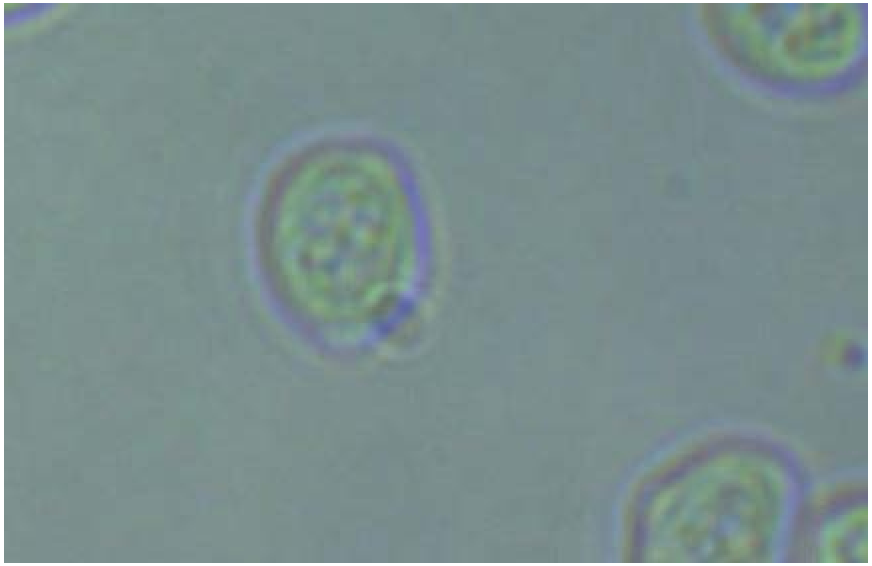
Benzene breathing in polystyrene

Characterization: Fluorescence



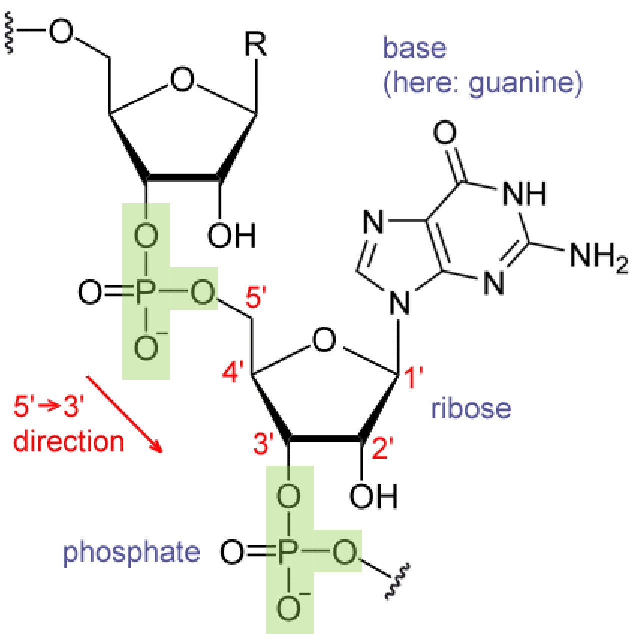


Fluorescence of dyed polystyrene bead

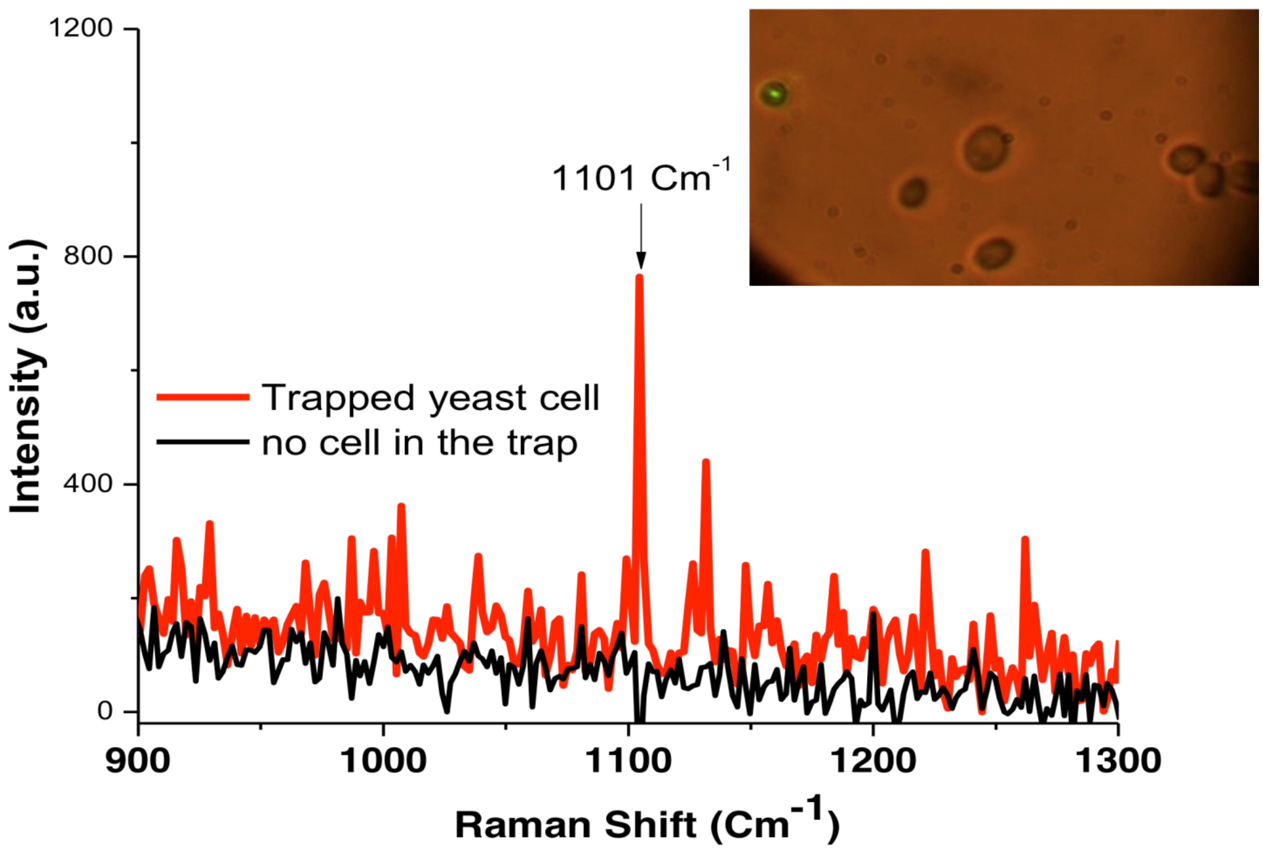


Results on yeast cell

Raman



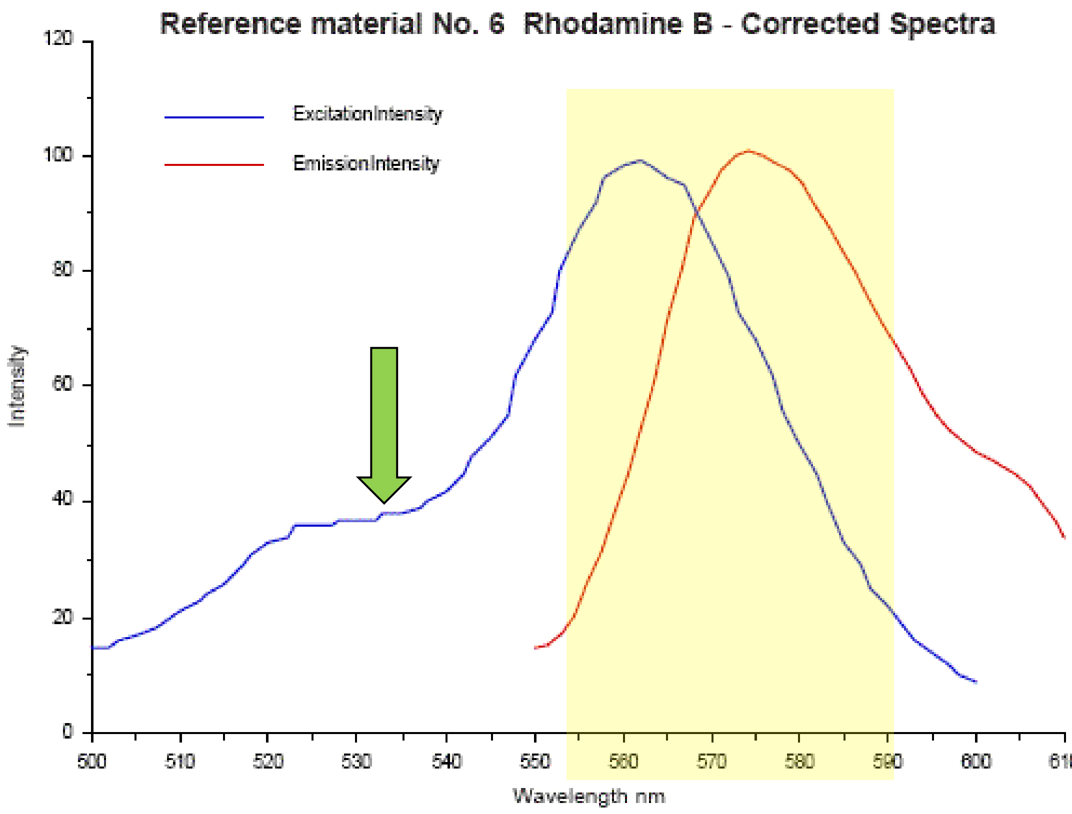
Daniel Ramsköld, Karolinska Institutet in Stockholm



P-O stretch in RNA molecule

Sample preparation

Yeast cells stained with Rhodamine B, hexyl ester, suspended in a 10mM Hepes buffer with 5% glucose for **mitochondria tagging**



http://www.chemistry.nmsu.edu/Instrumentation/fluorescence_Std_4.html

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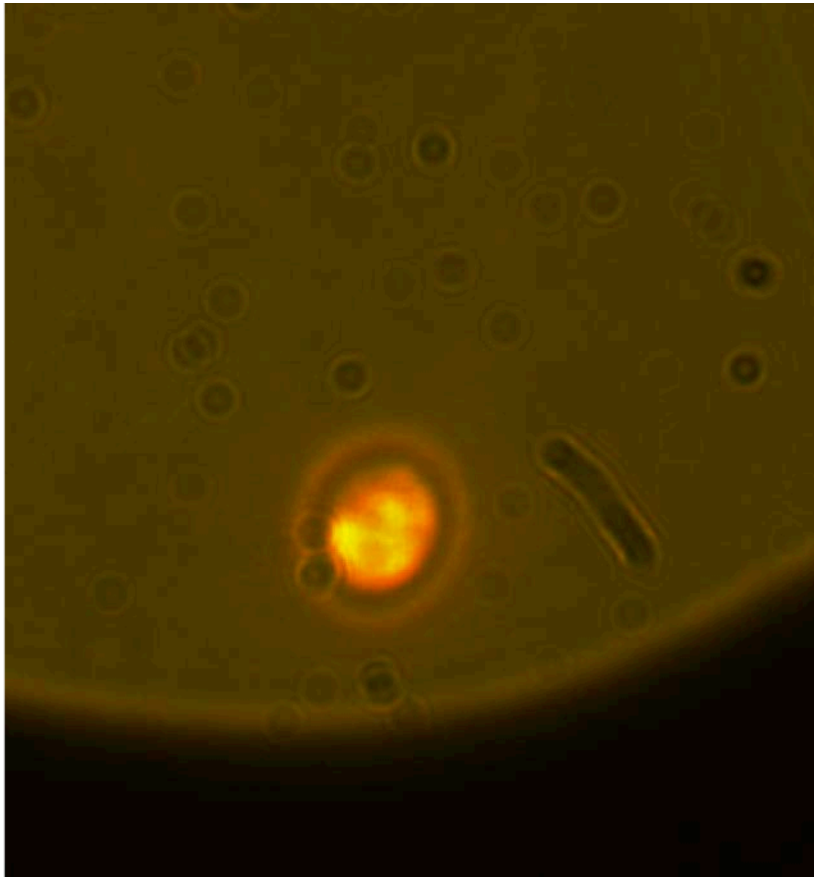
Fluorescence imaging



Imaging mitochondria in yeast cell

Only ~3 mitochondria: anaerobic character of the cell confirmed

Conclusion

Implementation of **vibrational and electronic spectroscopy** in optical tweezers

Imaging capabilities

Towards implementation of additional spectroscopic diagnostics

Acknowledgments

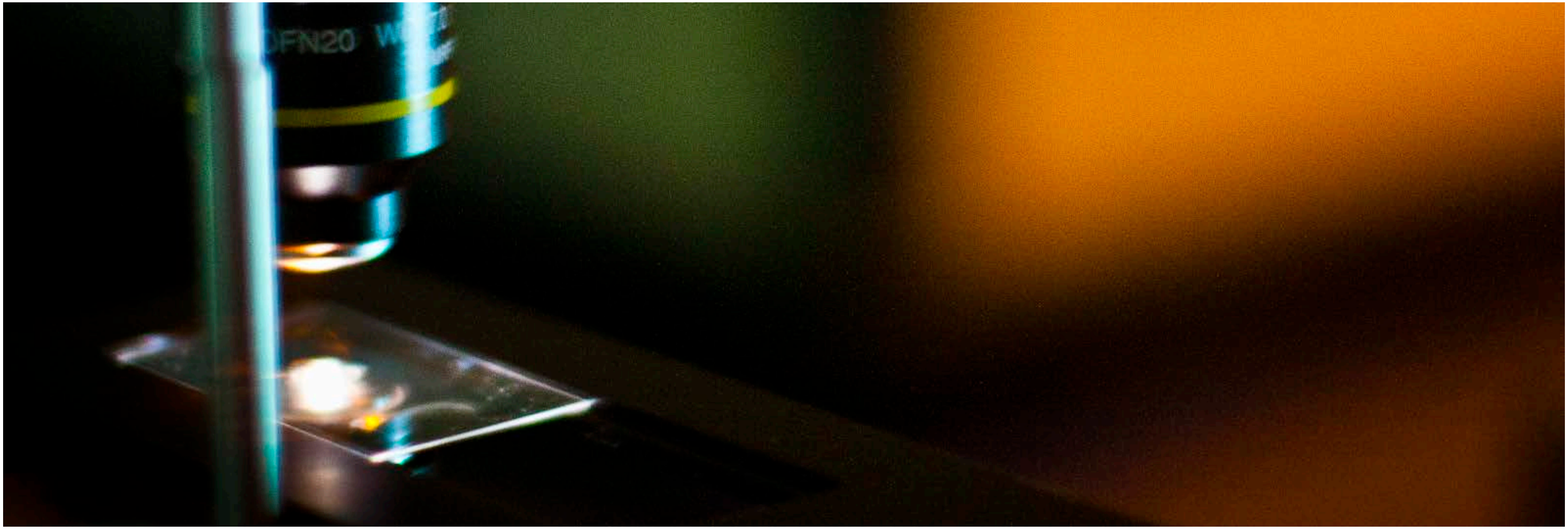
Funding

THORLABS

State of Florida

Industrial Affiliates

THANK YOU



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