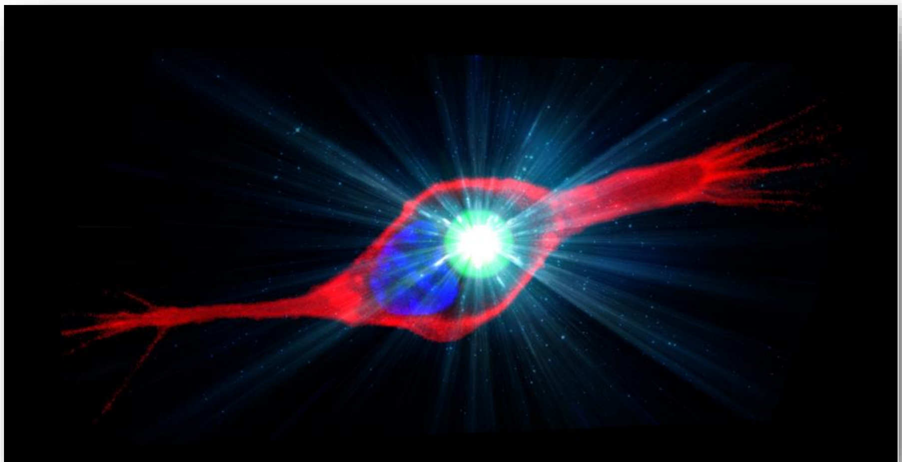
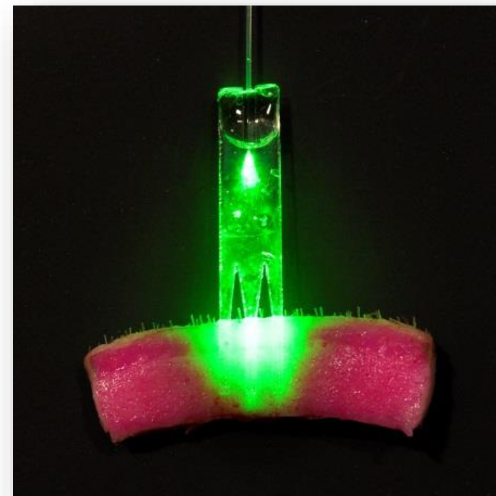
Live lasers and biocompatible optical fibers





Matjaž Humar

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http://www.matjazhumar.com

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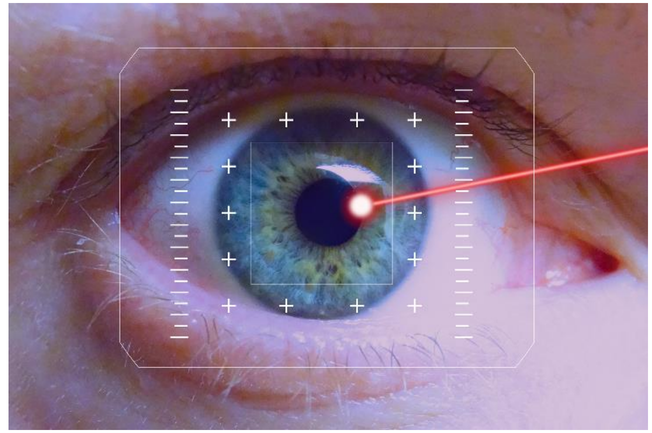
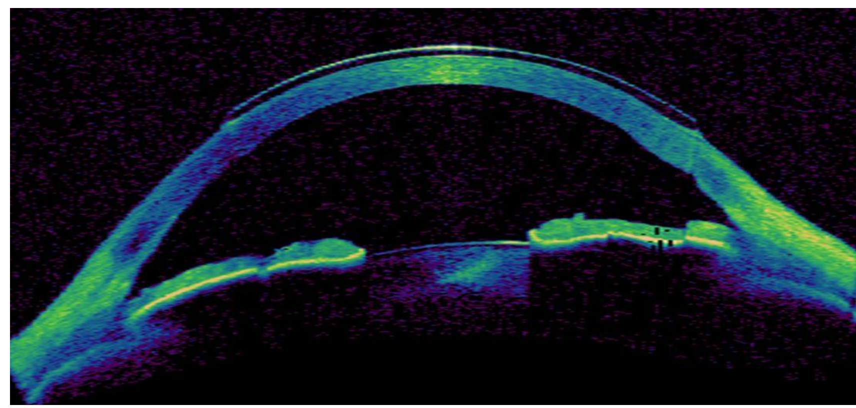
Use of lasers in medicine





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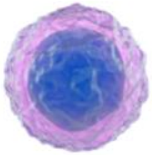
Bio-integrated photonics

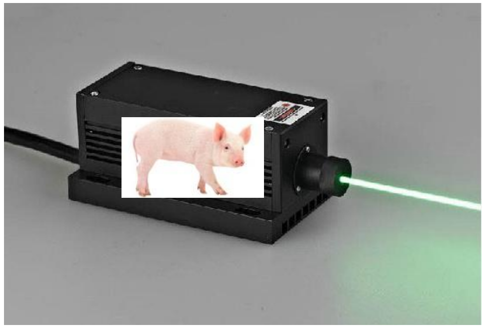


Biophotonic device

Treatments, diagnostics

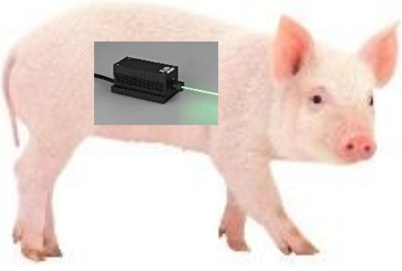
Biological system

Biosystem as part of the device



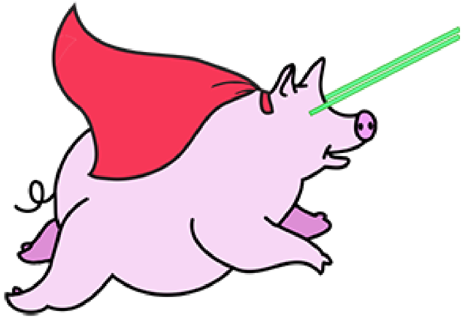
Example: cell inside a laser

Device inside a biosystem



Example: optical waveguide in tissue

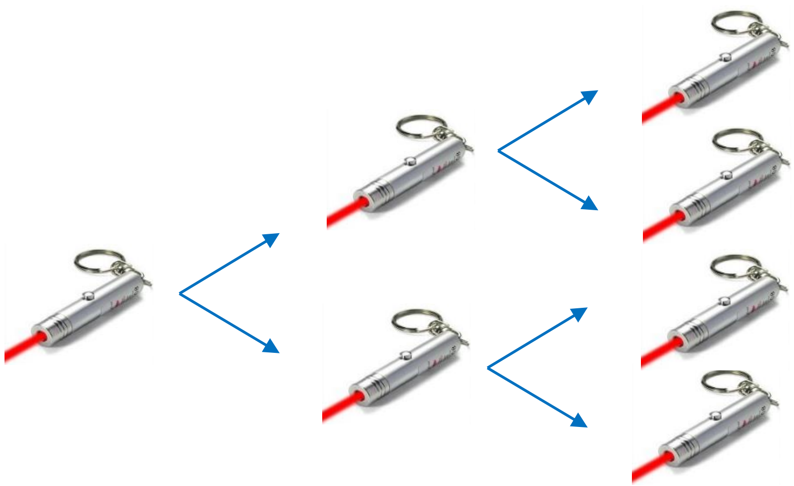
Device = Biosystem



Example: cell as laser

Why bio-integrated photonics

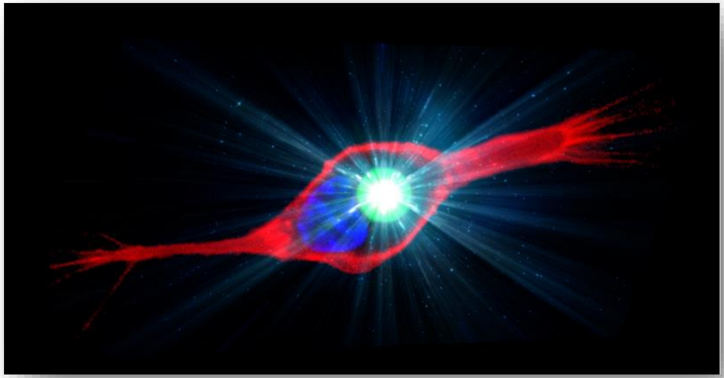
- Better coupling between the device and the biological system
  - Better sensors
  - Targeted medical treatments
- Biocompatibility and biodegradability
- Miniaturization
- Self-reproduction, self-assembly, evolution, adaptation, self-healing, scalability
- New human-robot interfaces (Cyborg)



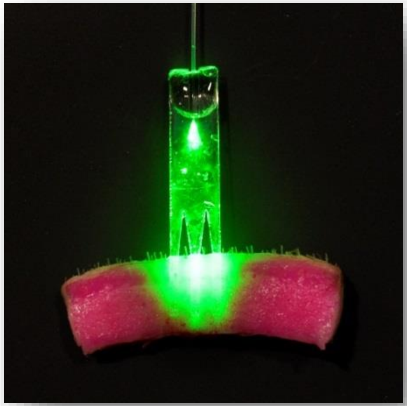
M. Humar et al., Toward biomaterial-based implantable photonic devices, Nanophotonics (2016).

Contents

- Cell lasers

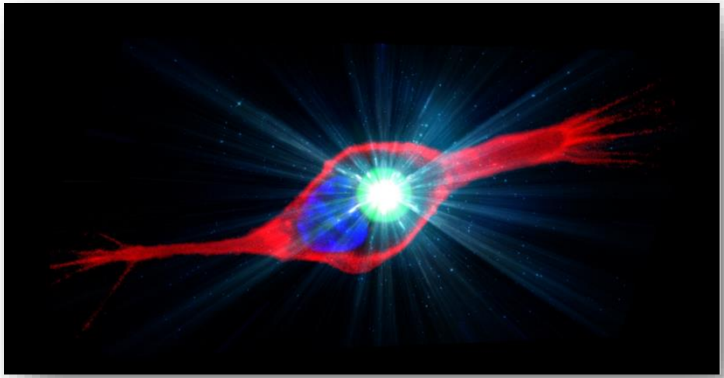


- Biocompatible optical waveguides

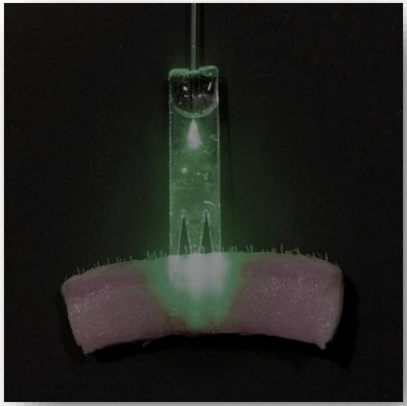


Contents

- Cell lasers



- Biocompatible optical waveguides



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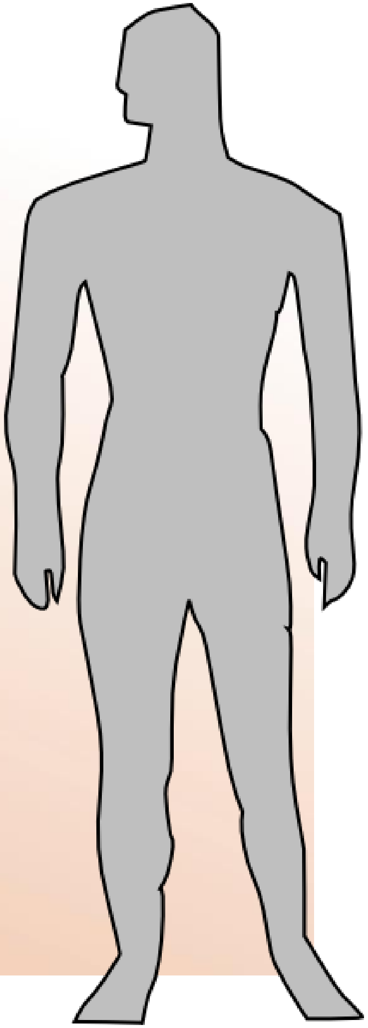
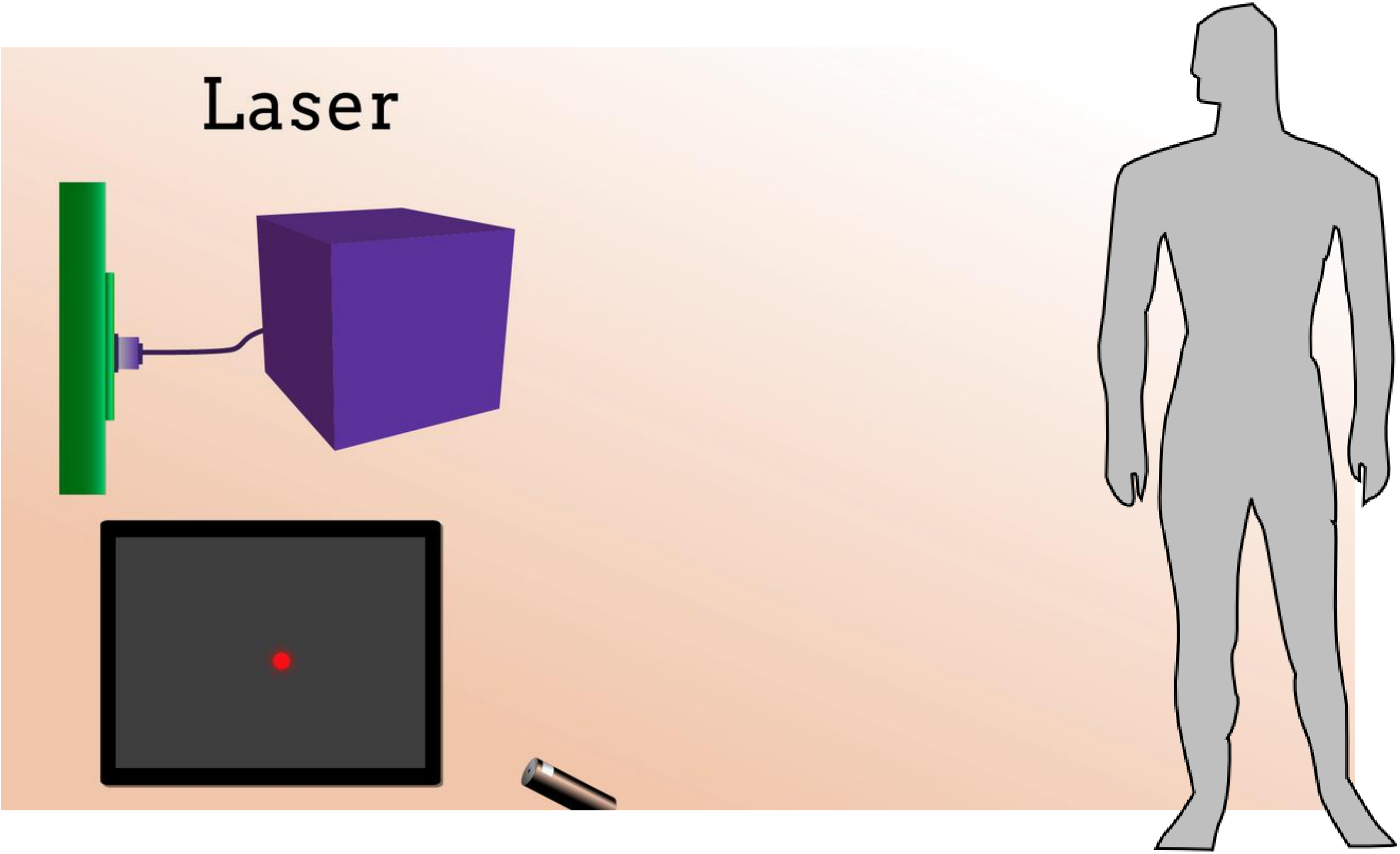
Laser inside biological tissues – paradigm shift

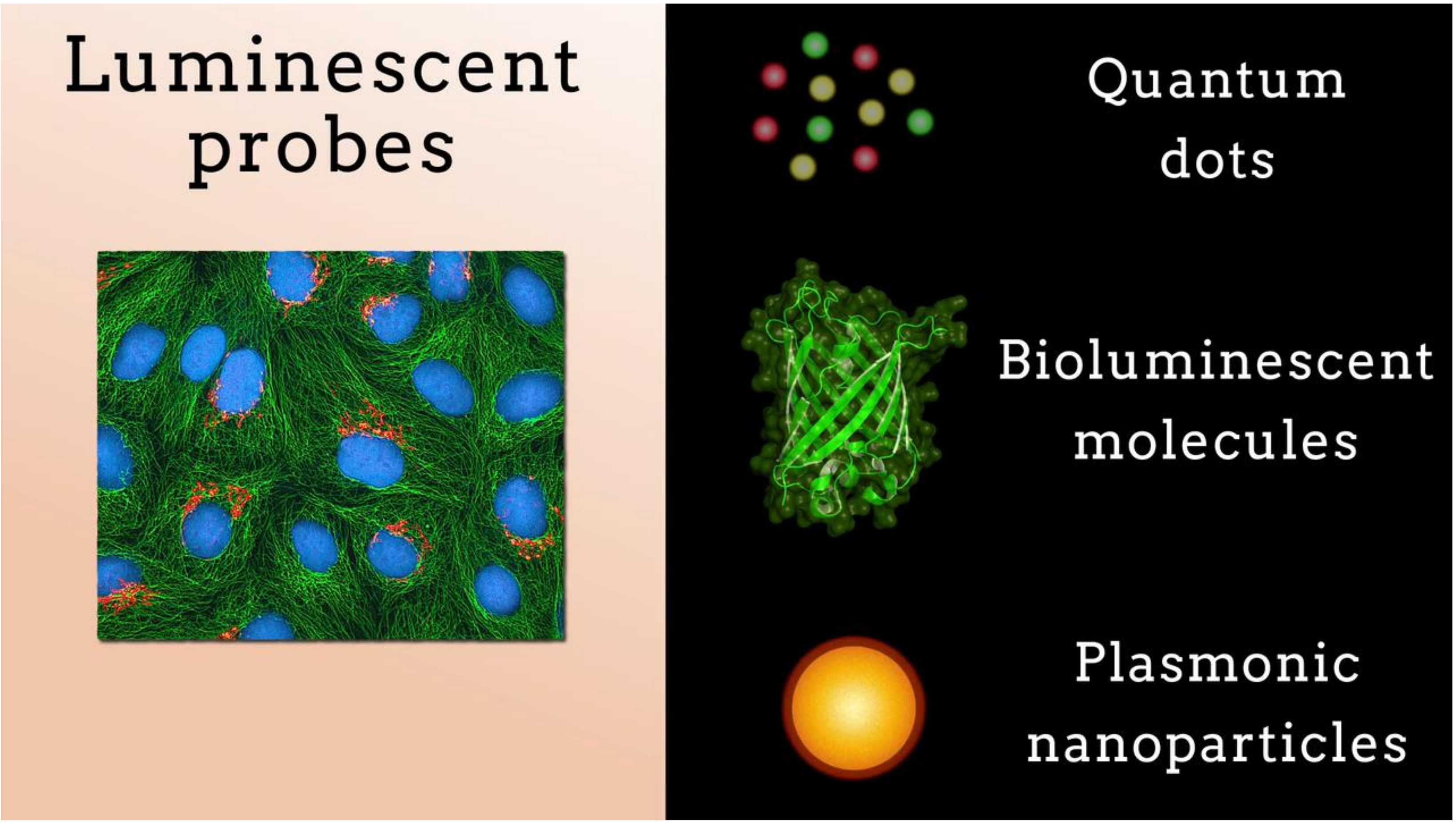
Laser



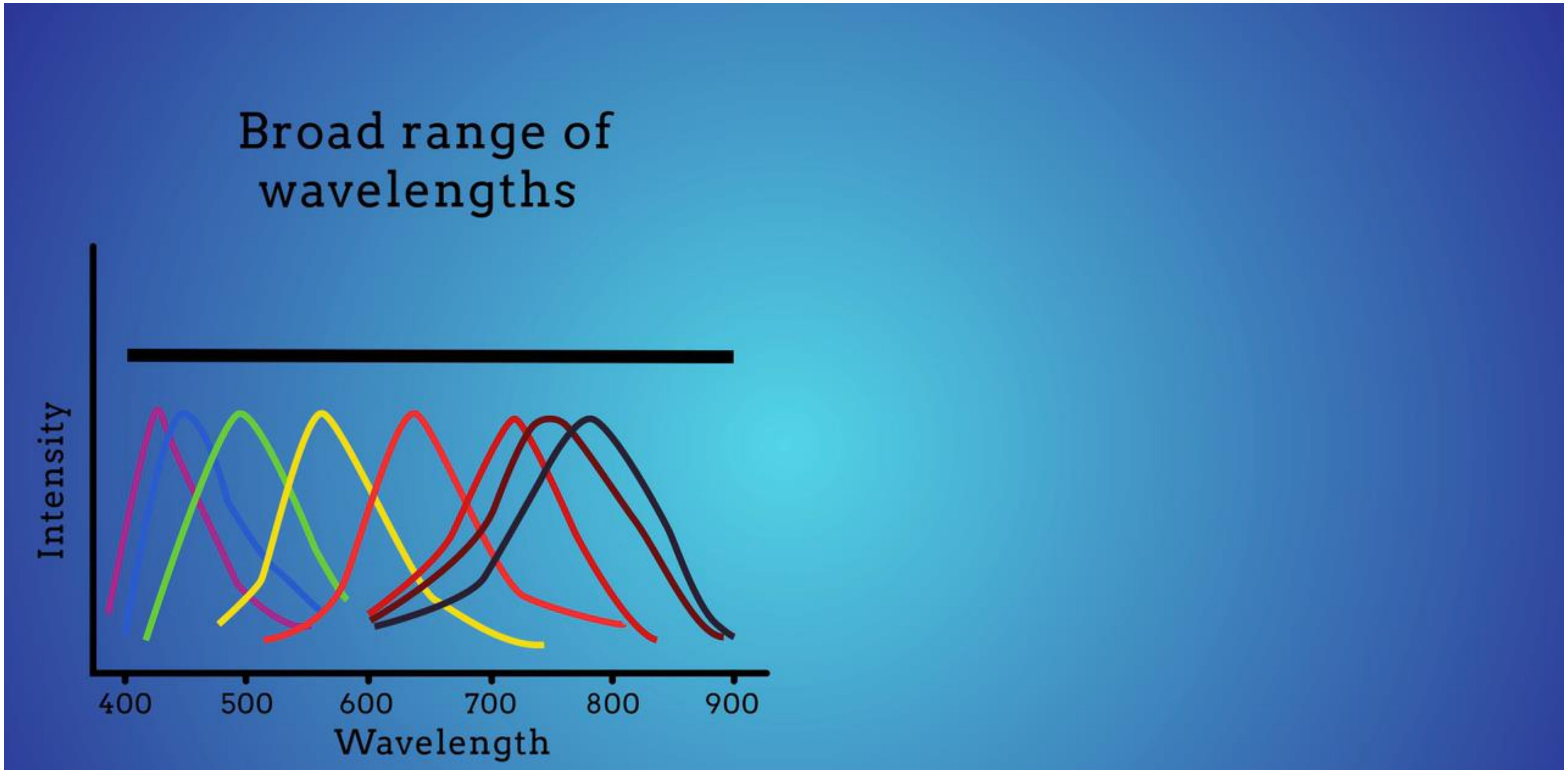
Laser inside biological tissues – paradigm shift

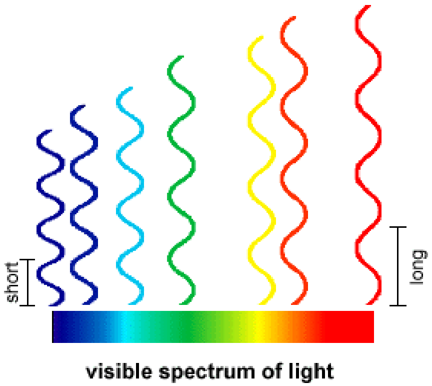


Fluorescent probes vs lasers

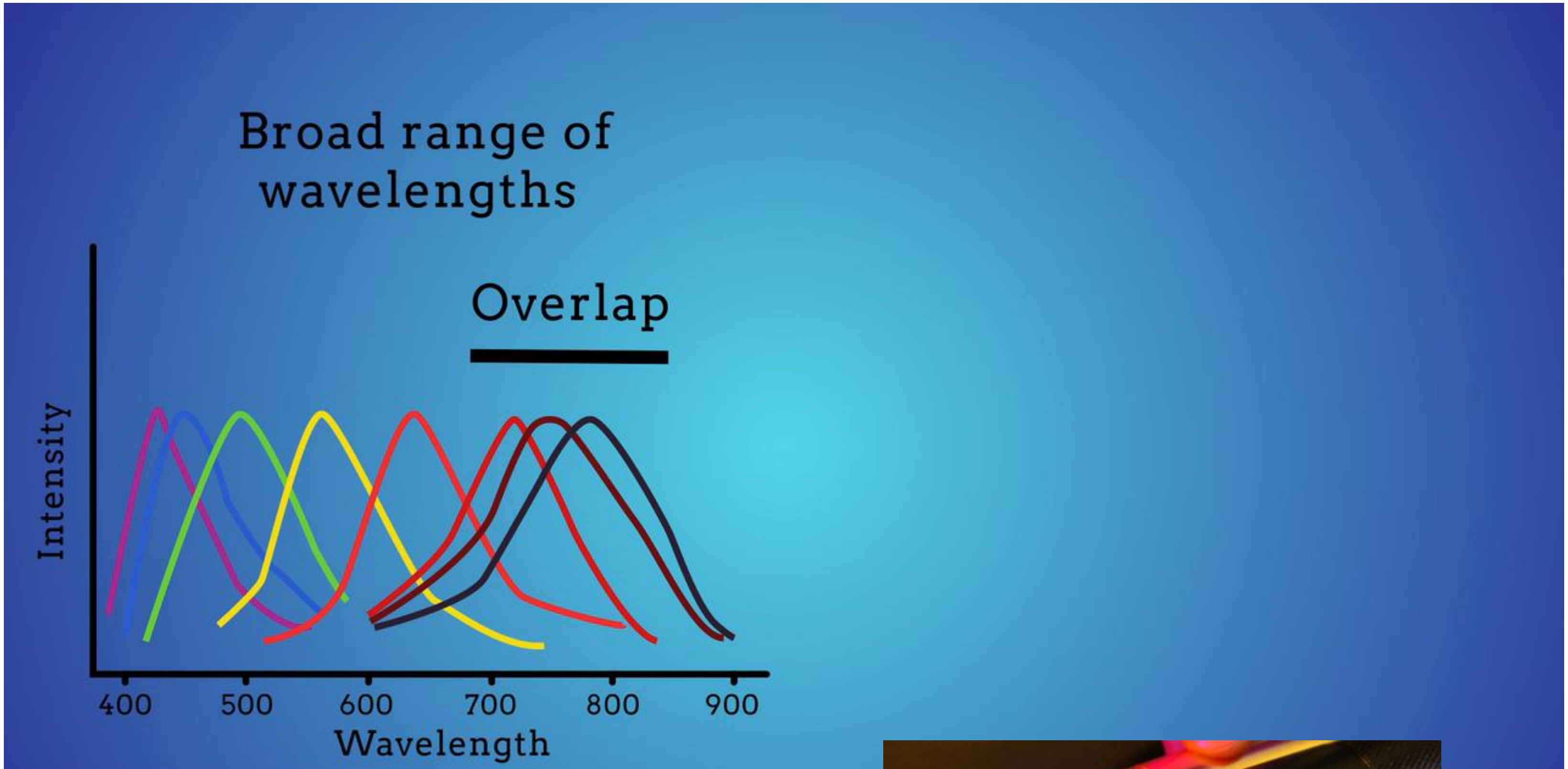


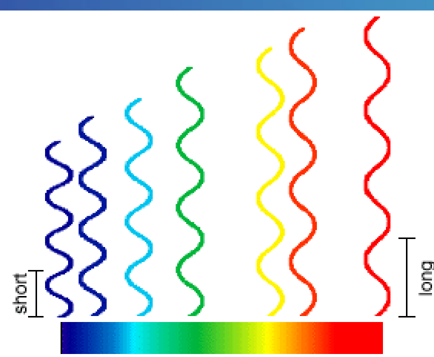
Fluorescent probes vs lasers



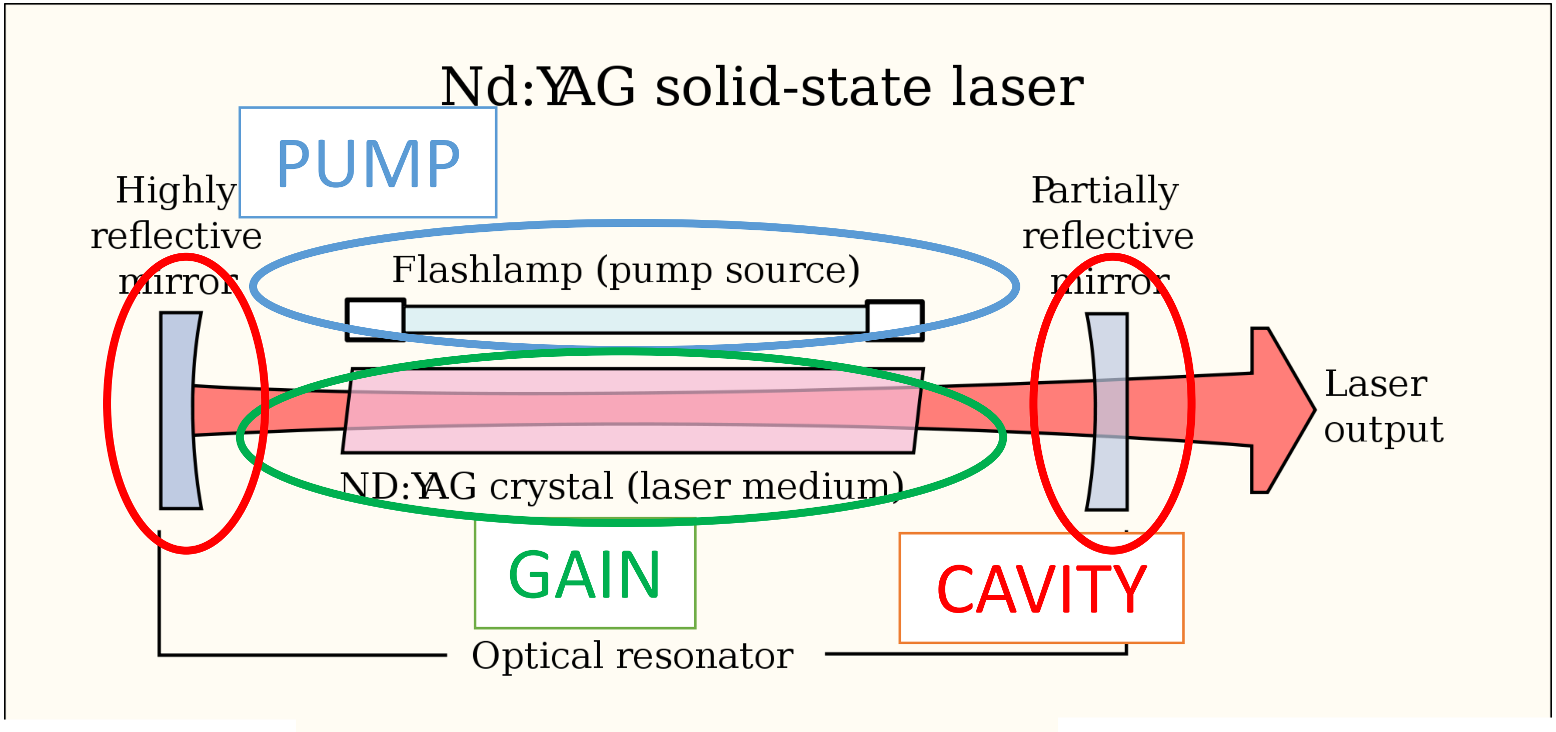


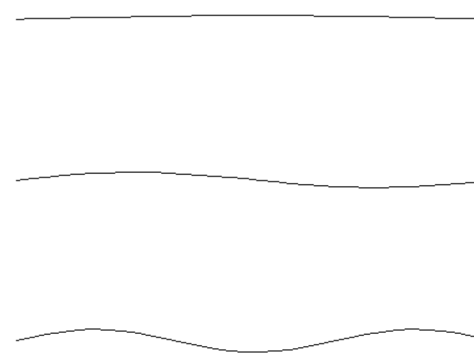
Fluorescent probes vs lasers

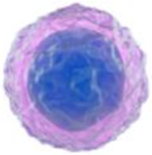




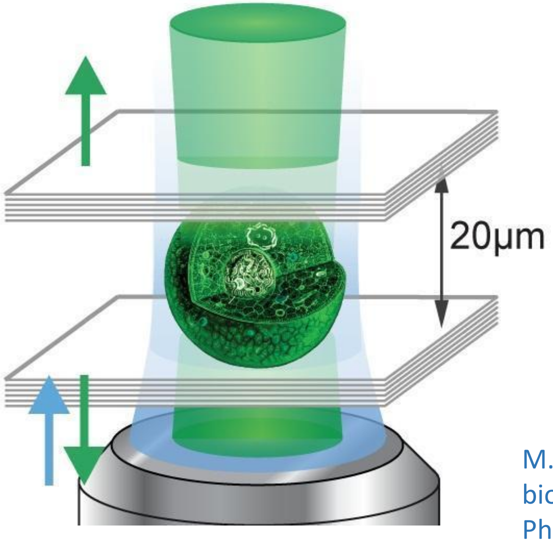
Lasers made completely out of biological materials



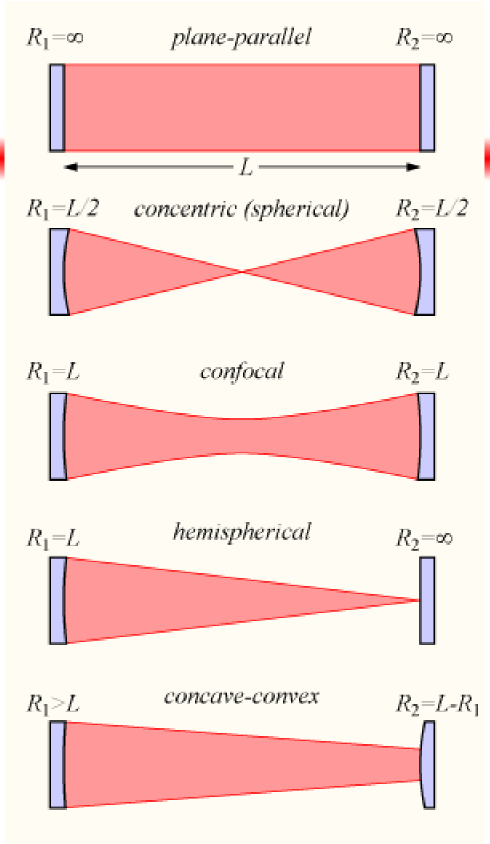




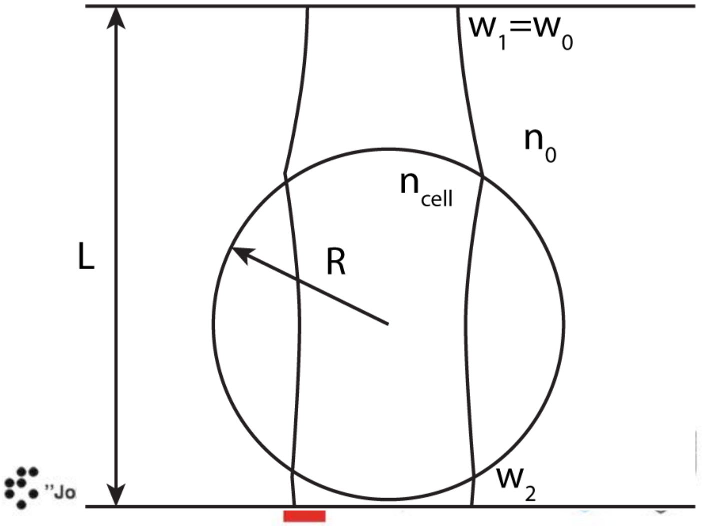
Cell inside a laser cavity

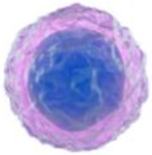


M. Gather, S.H. Yun, Single cell biological lasers. Nature Photonics 5, 406-410 (2011).



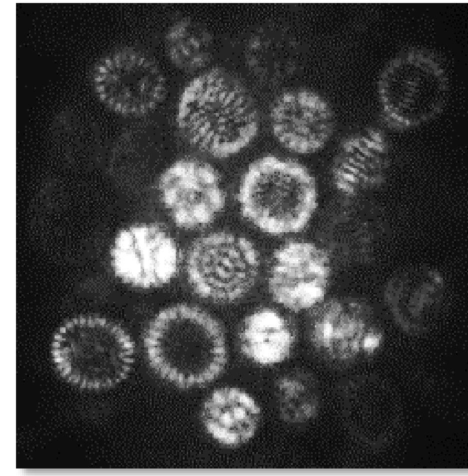
- Cell has a dual role
  - Lens – stabilizes the laser cavity
  - Gain materials– GFP (green fluorescent protein)

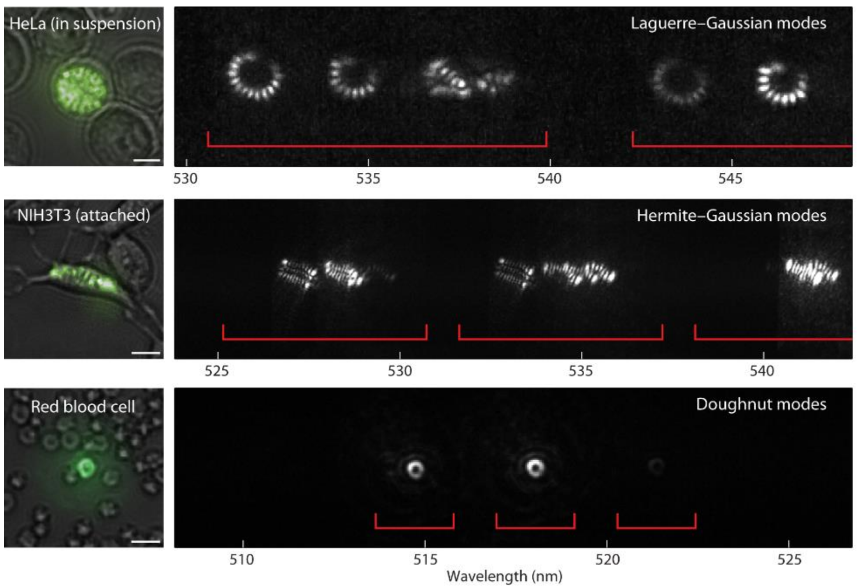


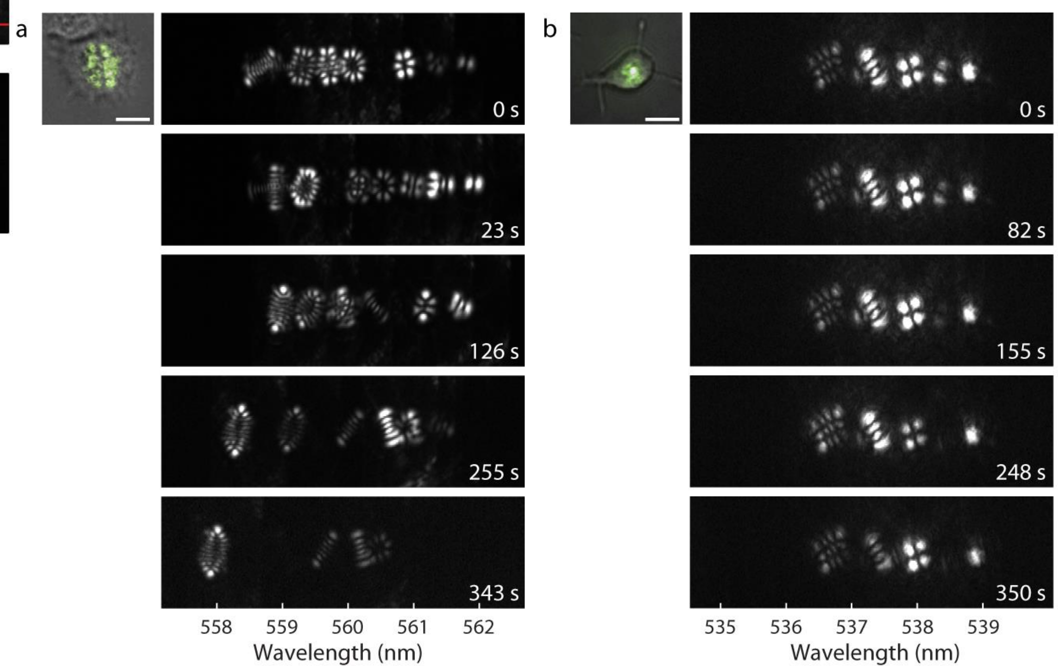


Laser modes emitted by the cell in the laser

- Characterization of cell types
- Extremely sensitive sensing

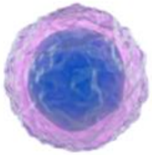




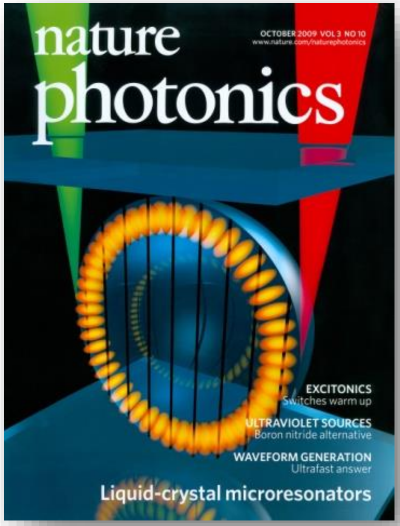


M. Humar, M. C. Gather, S.-H. Yun, Cellular dye lasers: Lasing thresholds and sensing in a planar resonator, Optics Express 23, 27865-27879 (2015).

S. Nizamoglu, K.-B. Lee, M. C Gather, K. S. Kim, M. Jeon, S. Kim, M. Humar, S.-H. Yun, A Simple Approach to Biological Single-Cell Lasers Via Intracellular Dyes, Advanced Optical Materials 3, 1197-1200 (2015).



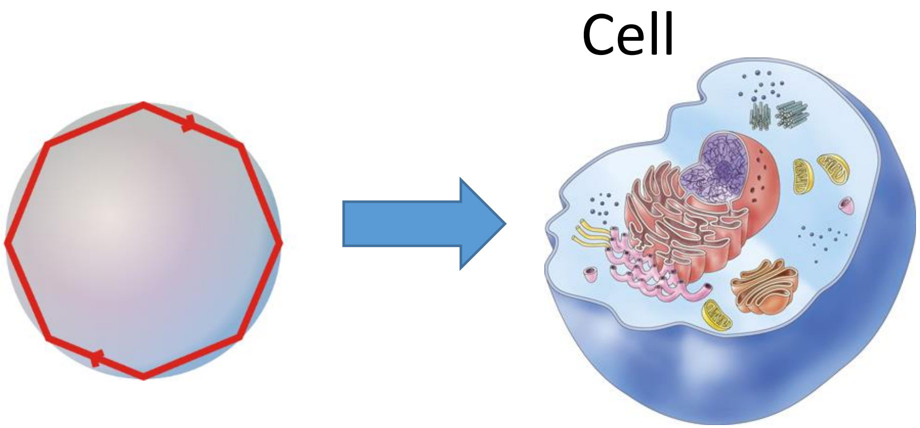
WGM cavities in cells

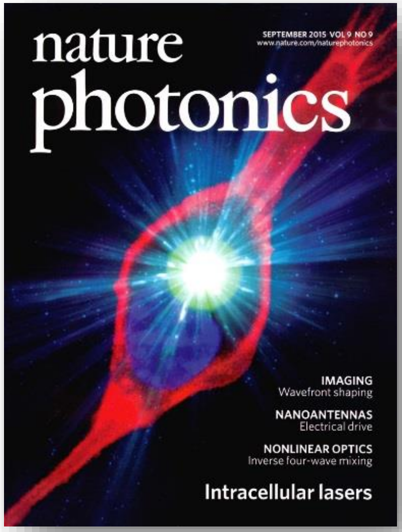


M. Humar, M. Ravnik, S. Pajk, and I. Muševič, Electrically tunable liquid crystal optical microresonators, Nat. Photonics 3, 595–600 (2009).

Whispering-gallery mode laser

- Light circulates due to total internal reflection
- Fluorescent dye as gain
- External laser pumping





M. Humar, S.-H. Yun, Intracellular Microlasers, Nature Photonics 9, 572-576 (2015).

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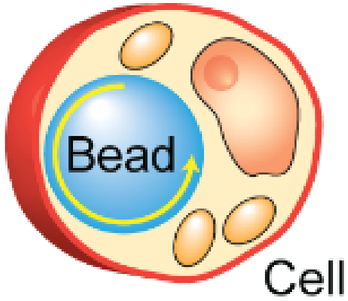
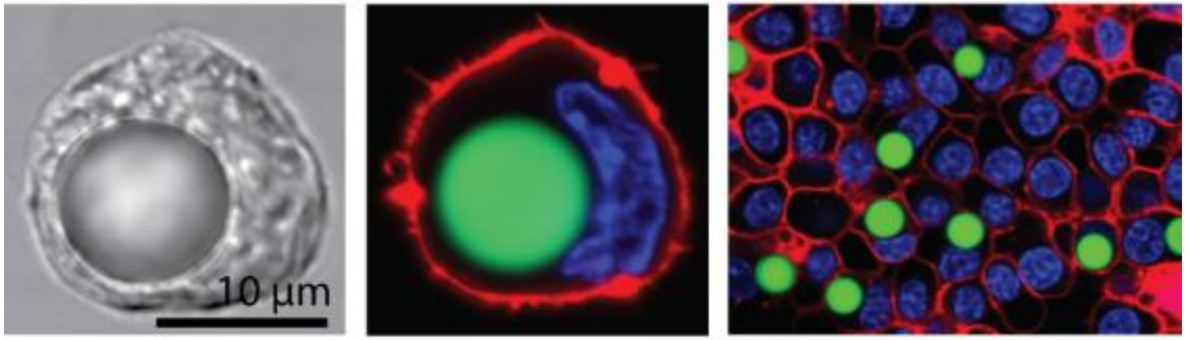
Solid beads – uptake by cells

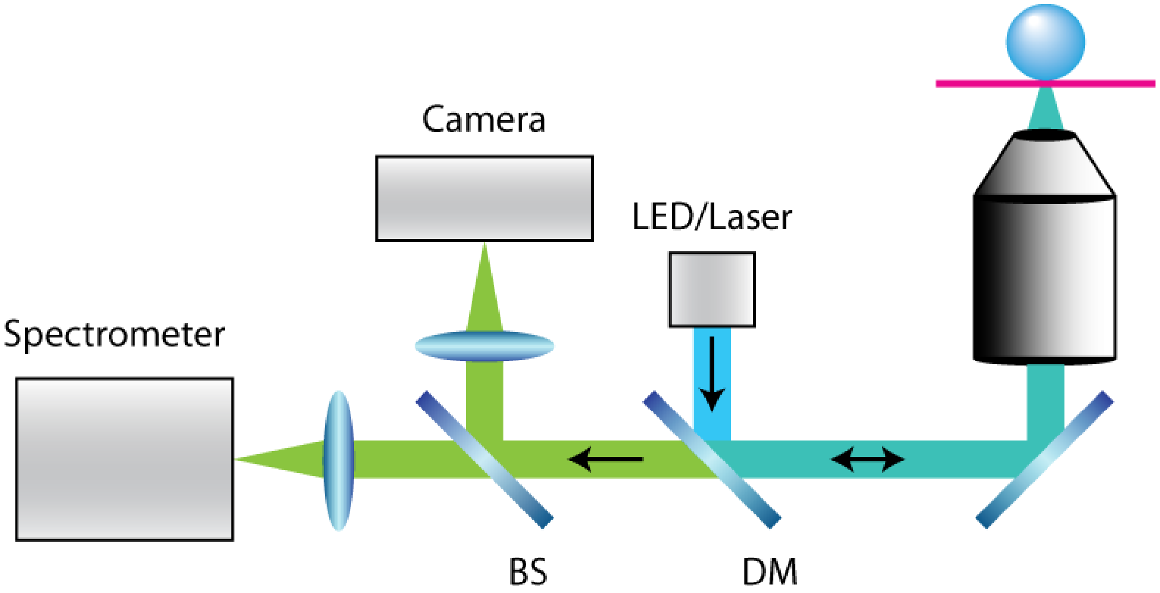
- Commercially available microbeads (1 – 15 µm)
- Materials: polystyrene, glass, $BaTiO_3$
- Engulfed by macrophage/non-macrophage cells by phagocytosis



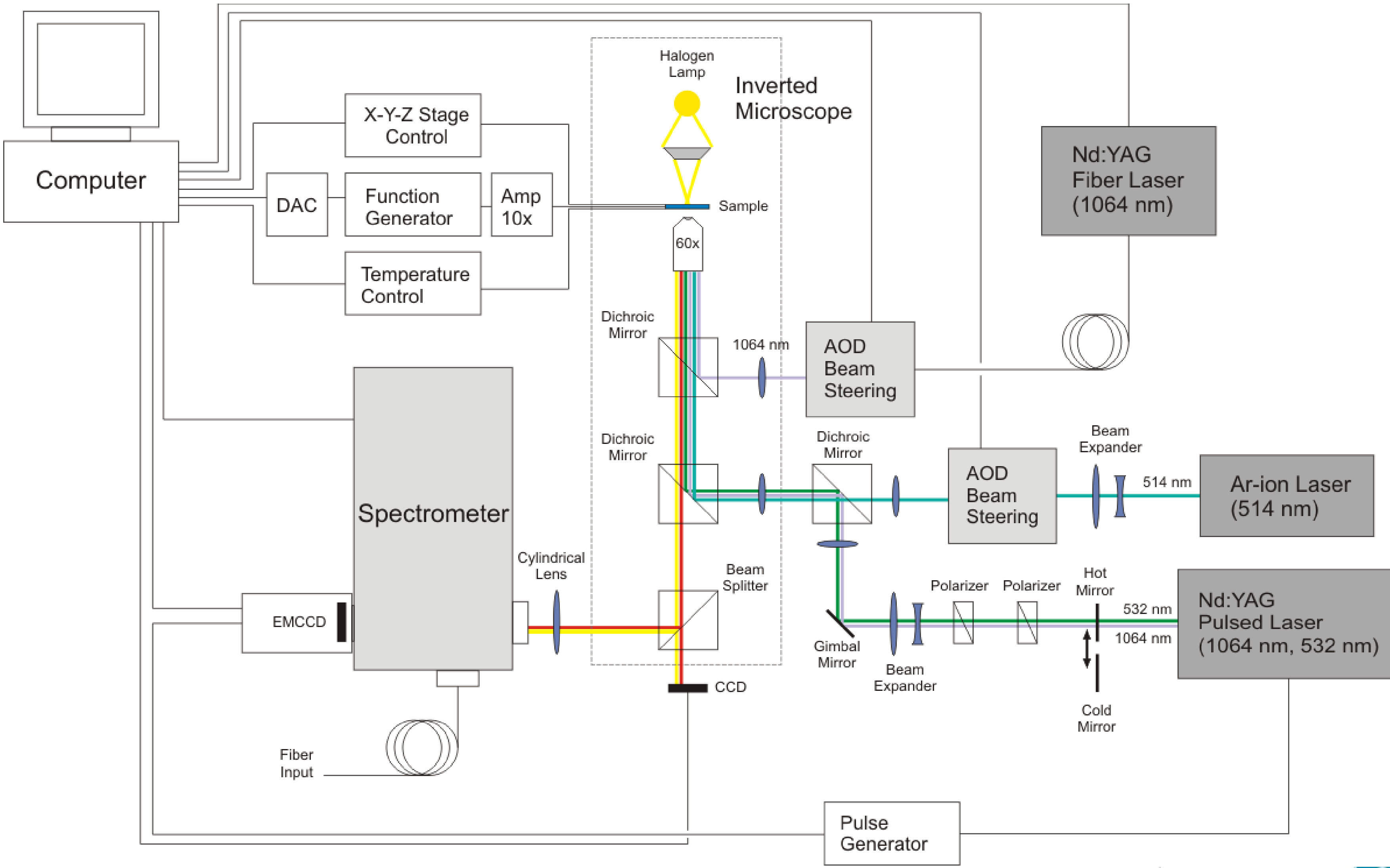


Optical system

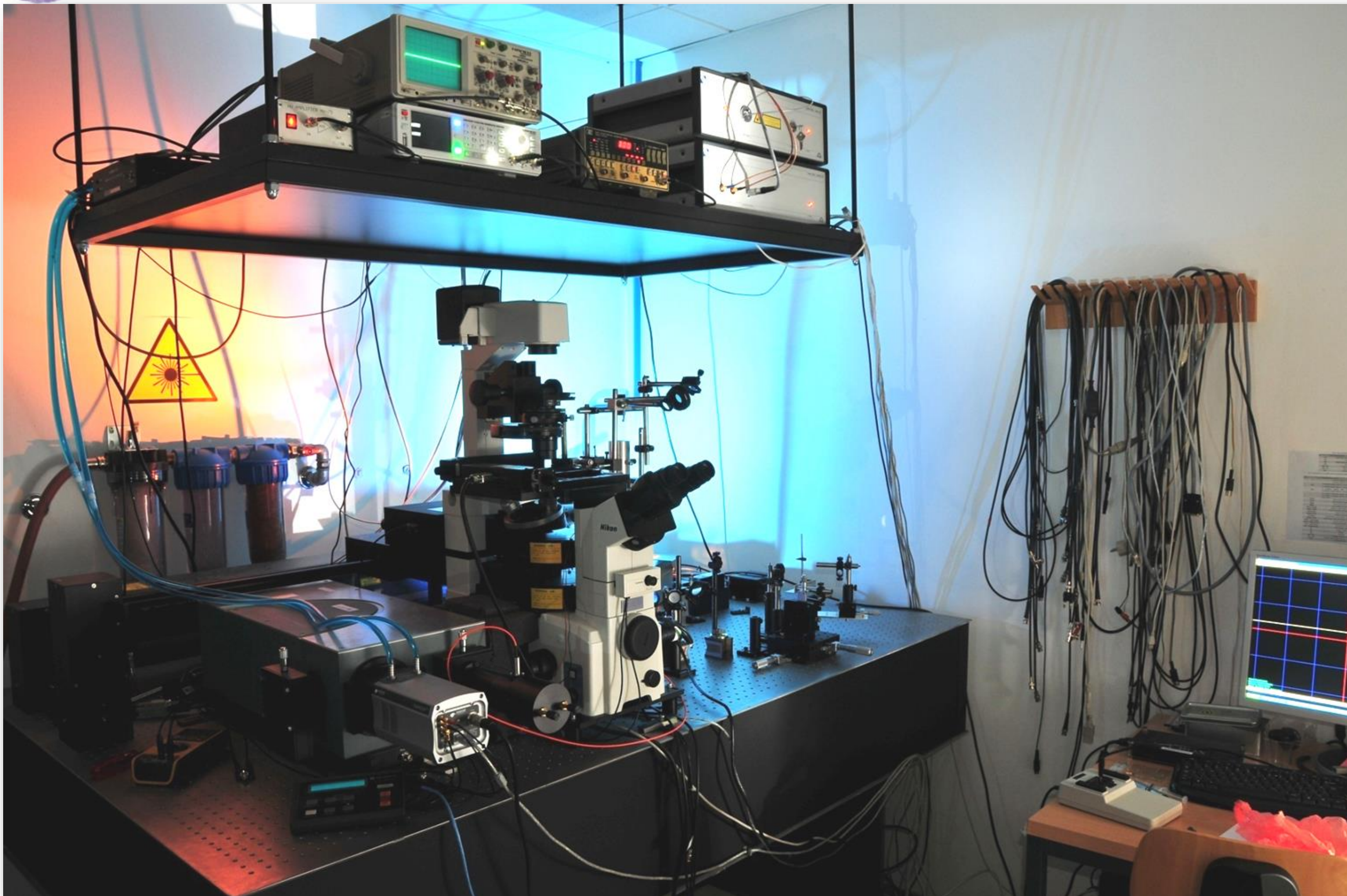
- External source of light
- Light from micro lasers is captured on
  - Camera
  - Spectrometer



Optical system

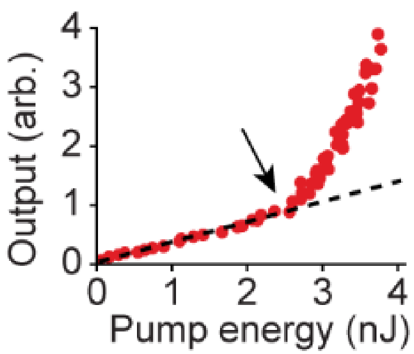


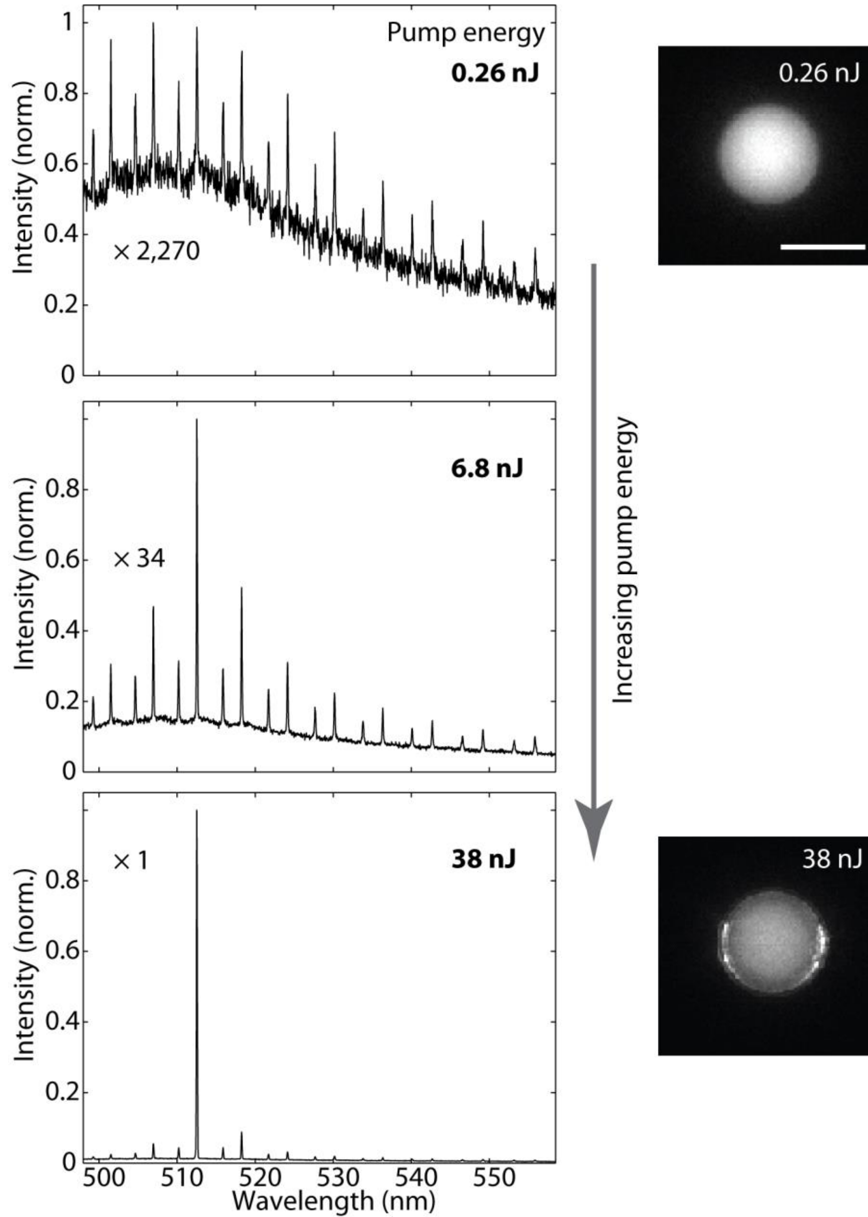
Optical system



Solid beads - lasing

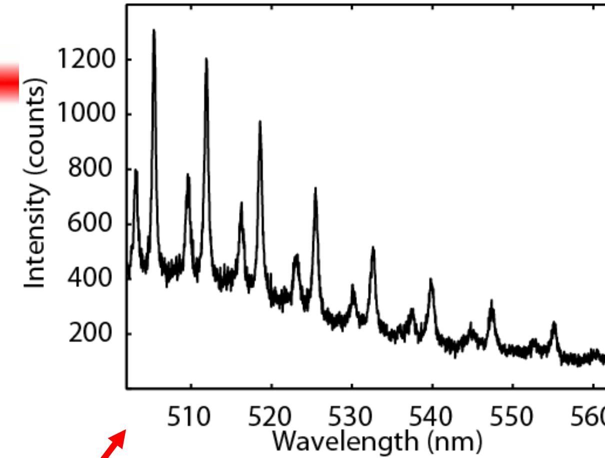
- Pumping with an external laser
- Sharp spectral peaks
- Whispering gallery modes
- High intensity above lasing threshold

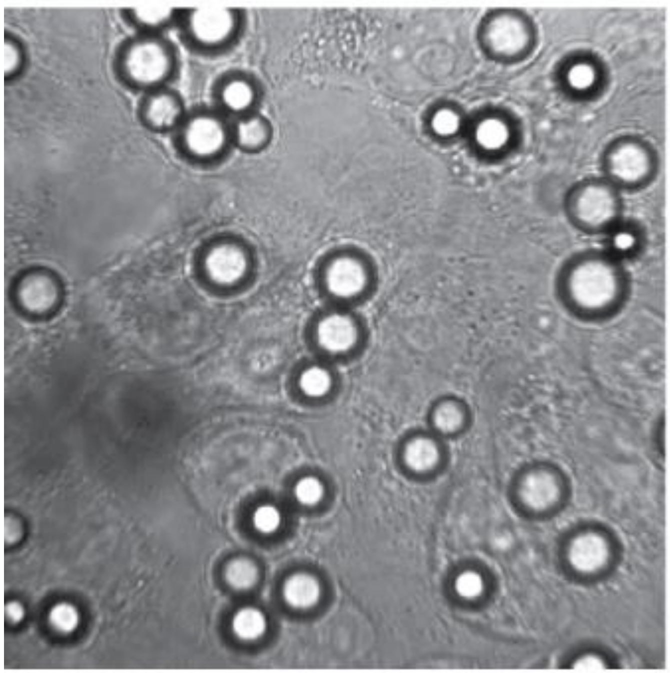


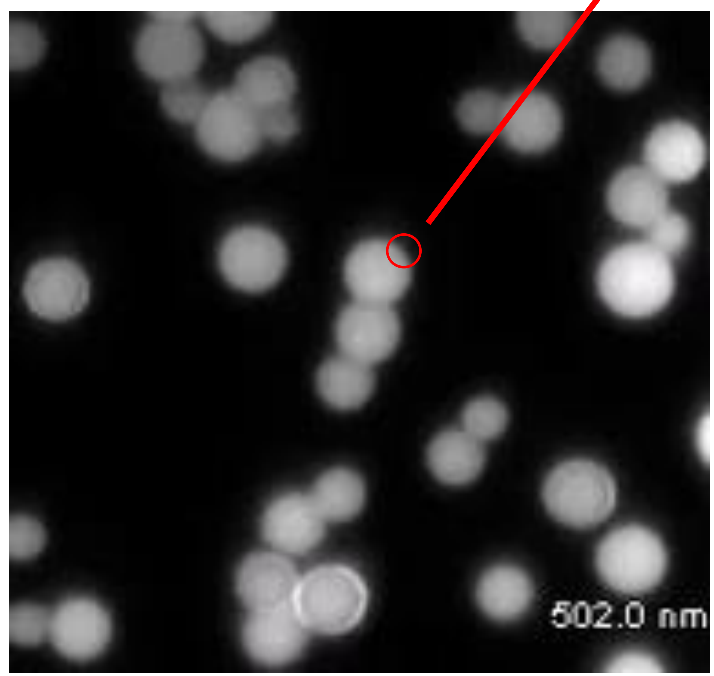


Cell tagging

- Hyperspectral confocal fluorescence image
- Blinking corresponds to WGMs





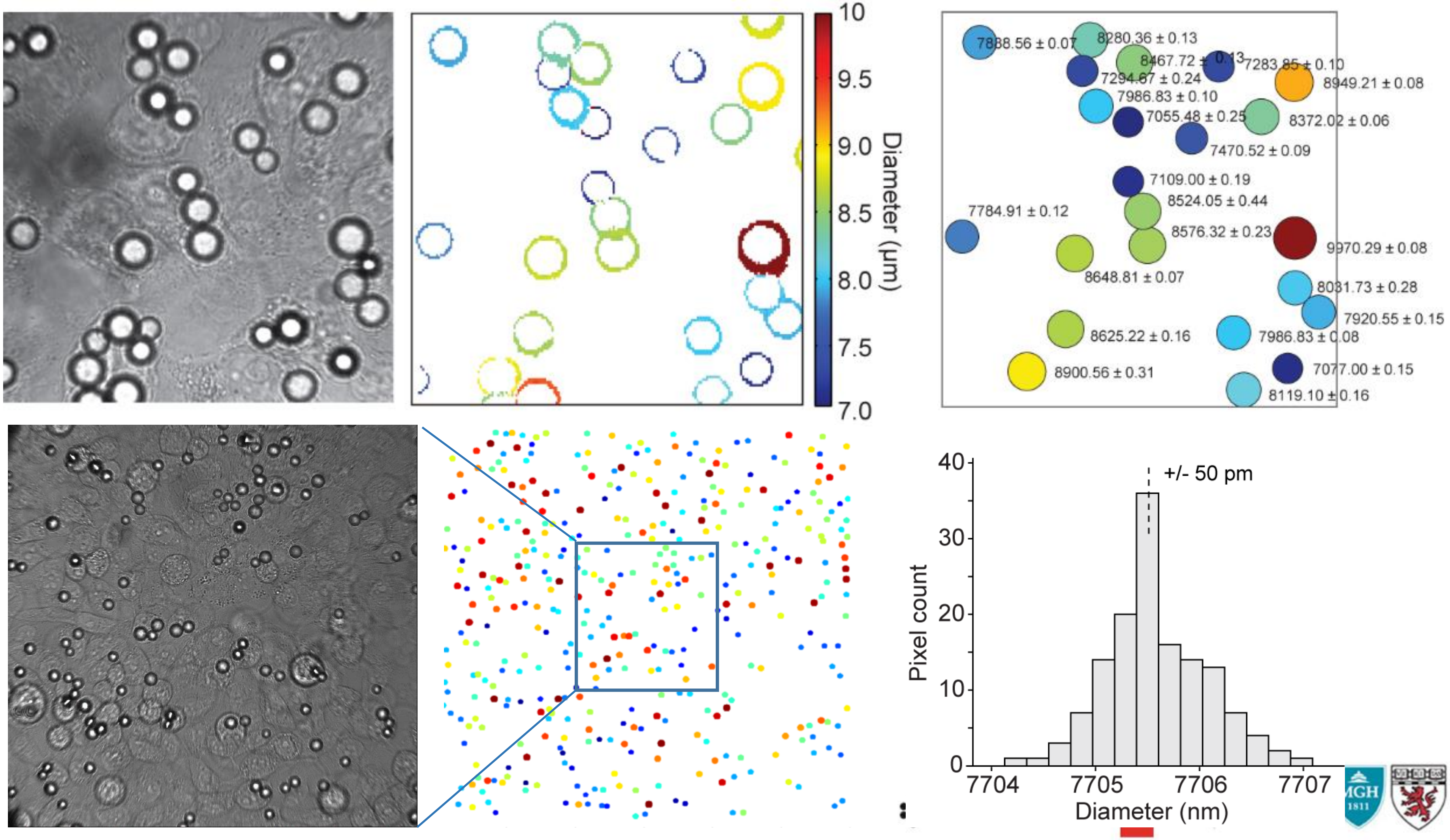


Fitting

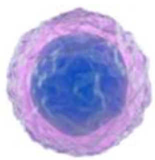
d = 8576.32 ± 0.23 nm

Hyperspectral imaging

- Calculate bead diameter
- Sensitivity: 50 pm / 7.7 µm = 6.5×10^{-6}



Cell tagging



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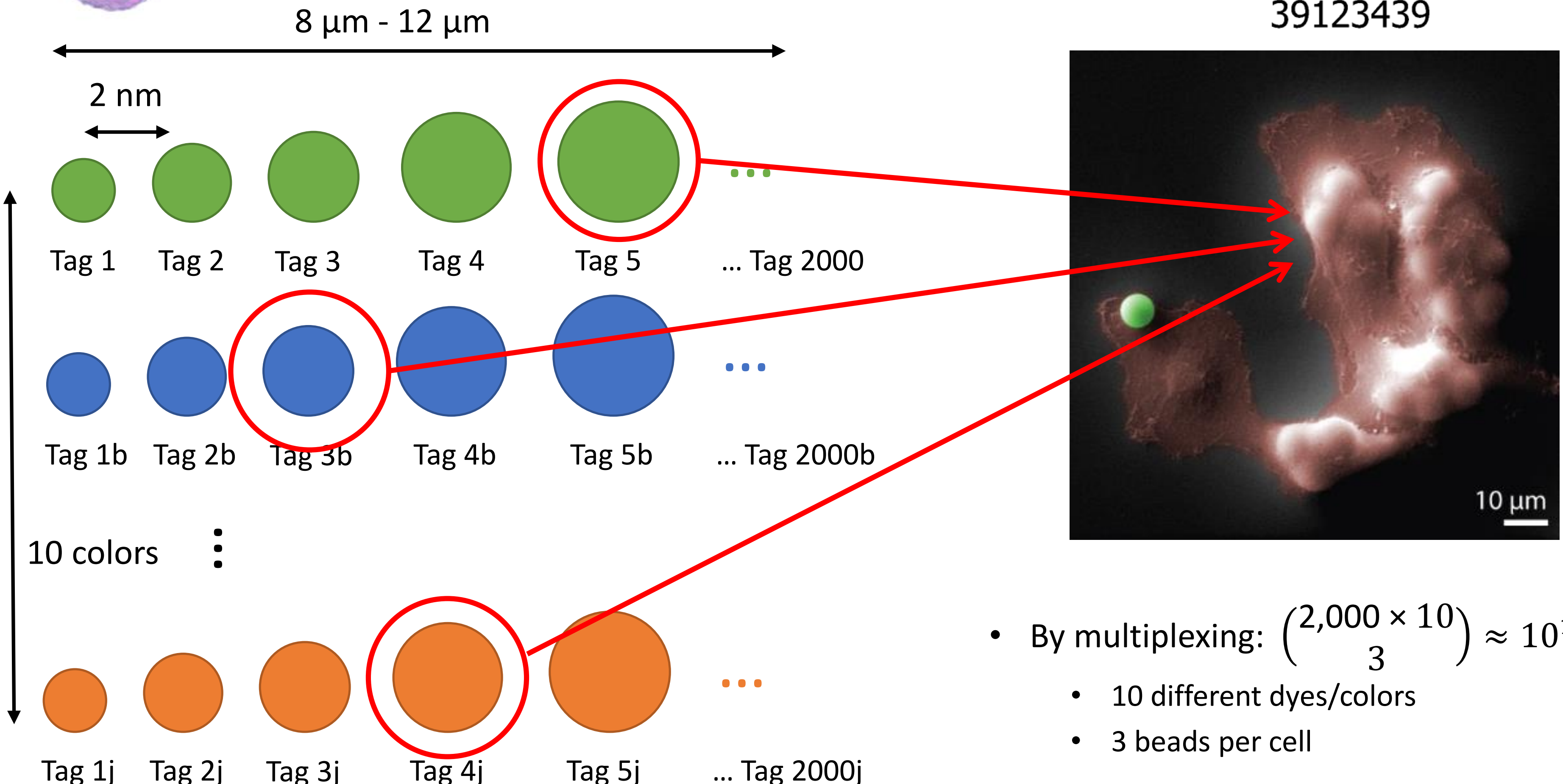

- By multiplexing: $\binom{2{,}000 \times 10}{3} \approx 10^{12}$
  - 10 different dyes/colors
  - 3 beads per cell
- On the same order as the number of cells in the human body

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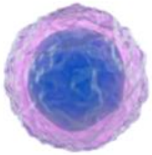
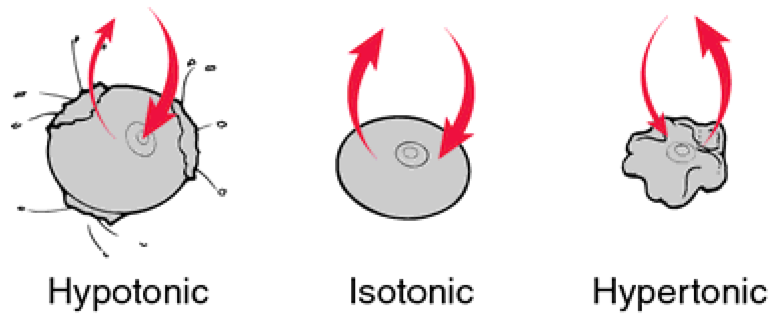
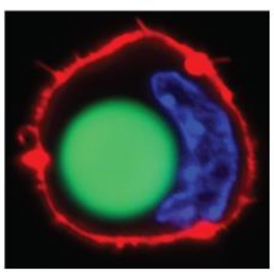


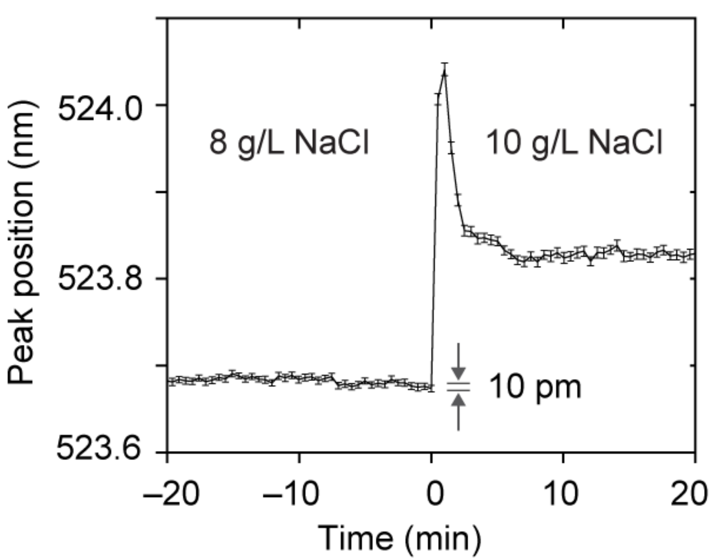
Intracellular sensing

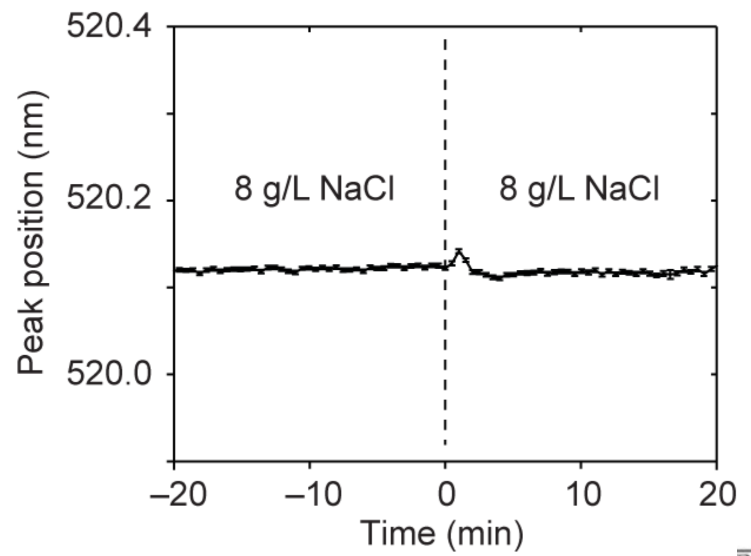
- Whispering gallery modes are sensitive to surrounding refractive index
- Proof of concept measurement
  - Increase of external osmolarity
  - Decrease in cell volume
  - Increase in refractive index
- We can measure change in refractive index down to 2.9×10^{-4} RIU



http://medical-dictionary.thefreedictionary.com

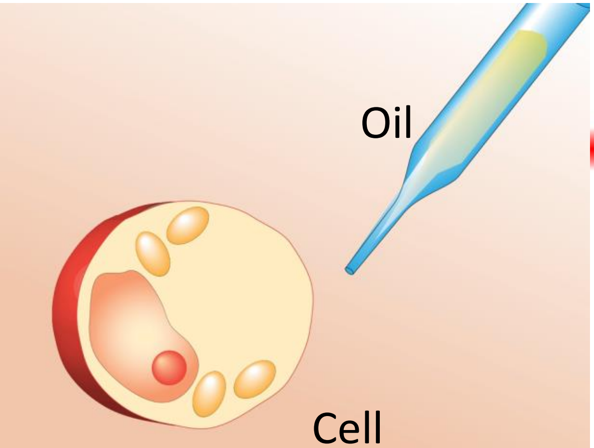


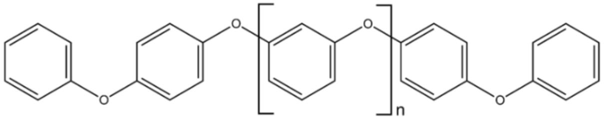


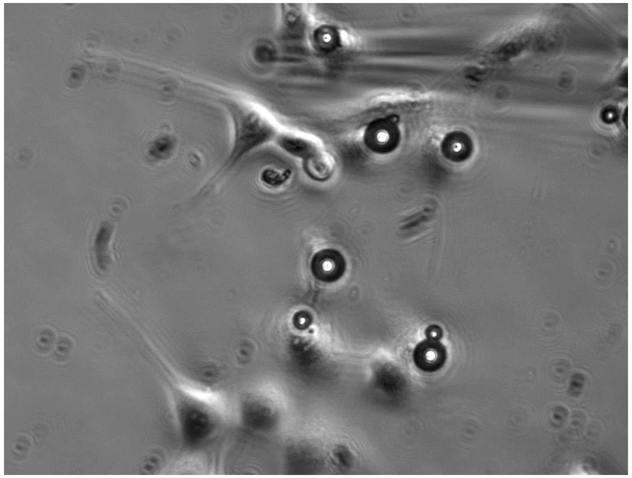


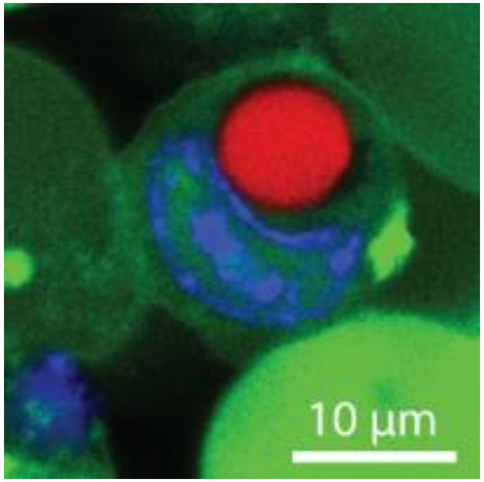
Injected high index oil

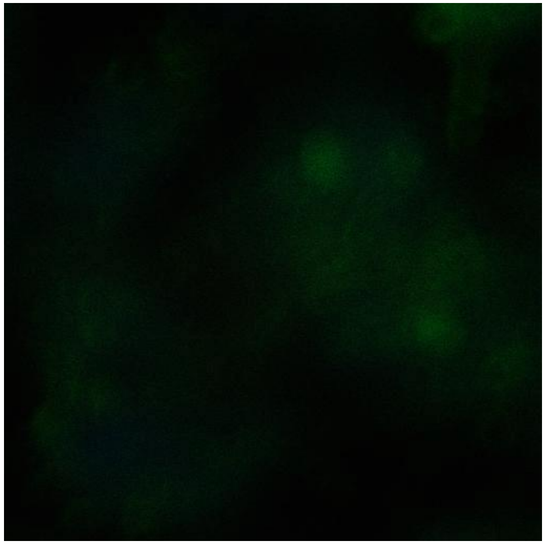
- Standard microinjection apparatus
  - Typically used to inject DNA
  - Glass micropipette with tip diameter ~1 μm
- Polyphenyl ether oil
  - High refractive index (n = 1.69 @ 500 nm)
  - Low viscosity, Insoluble in water, Nontoxic
  - Nile red fluorescent dye, 5 mM











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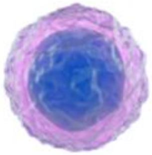
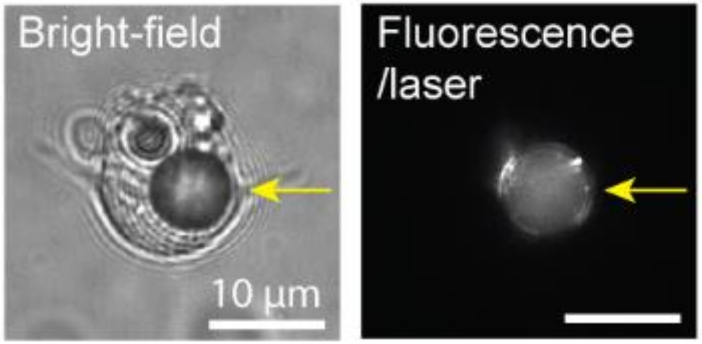
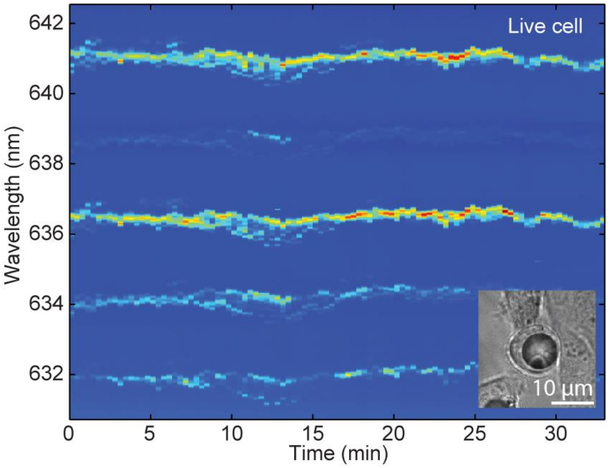
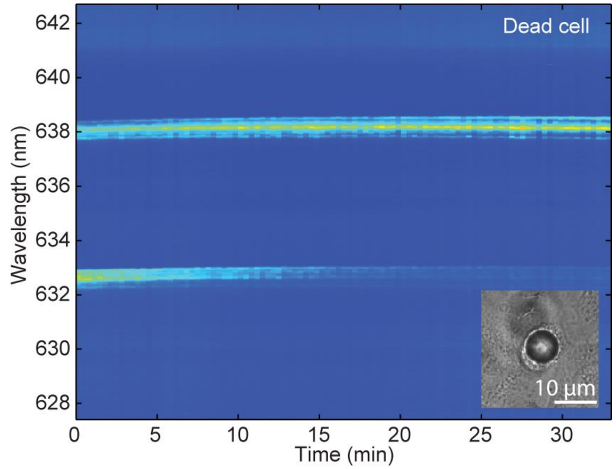


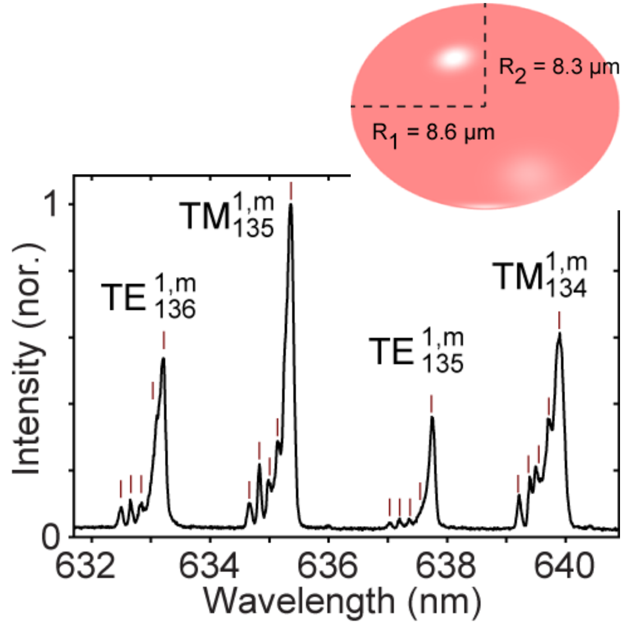
Injected high index oil - lasing

- Droplet deformation causes splitting of spectral lines
    - Enables approximate force calculation
    - Real time intracellular force tracking
- Force fluctuation sensitivity: 40 pN/µm^2 (40 Pa)









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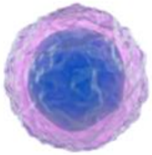
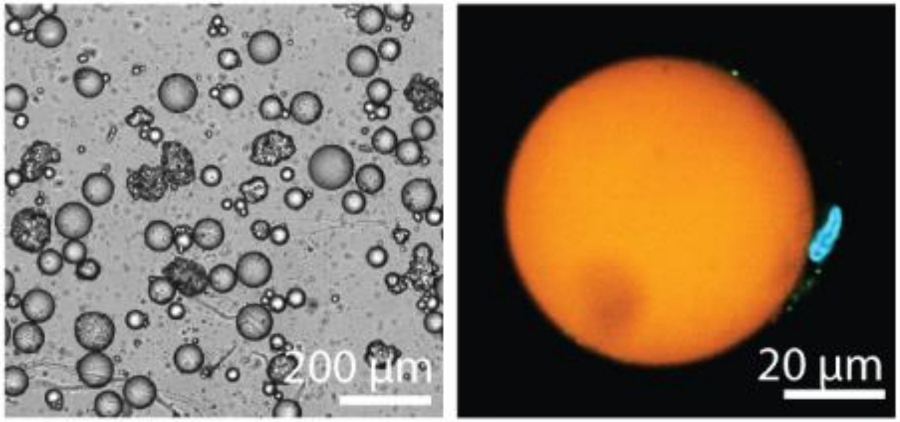
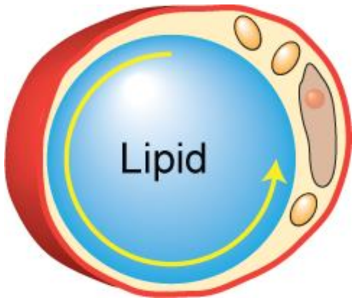
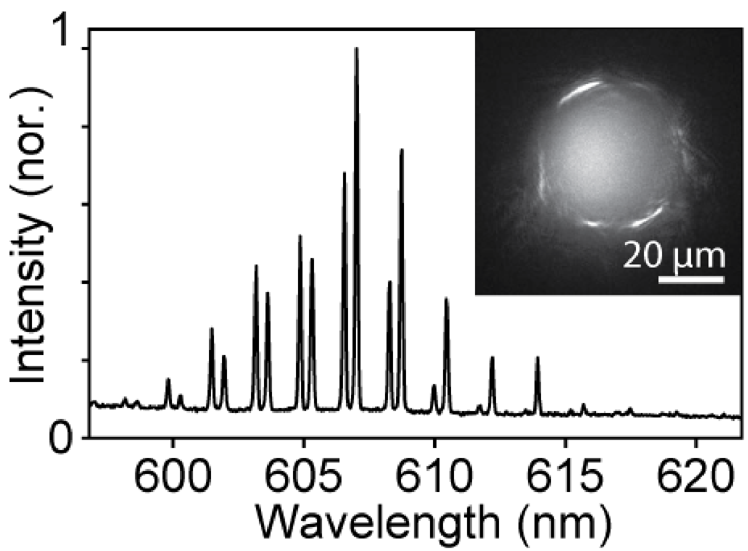


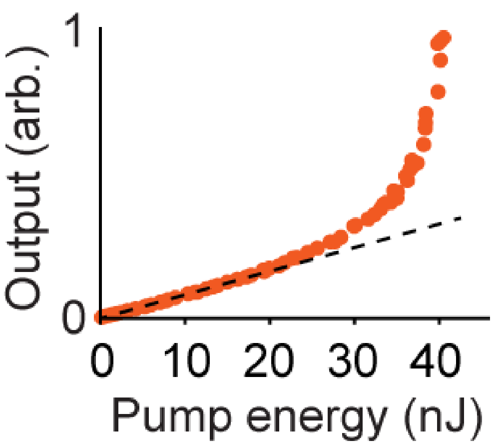
Adipocytes – fat tissue cells lasing

- Adipocyte cells naturally contain large lipid droplets
- Adipocytes were harvested by collagenase digestion from porcine subcutaneous fat tissue
- Stained with nile red
- Pumped with external laser



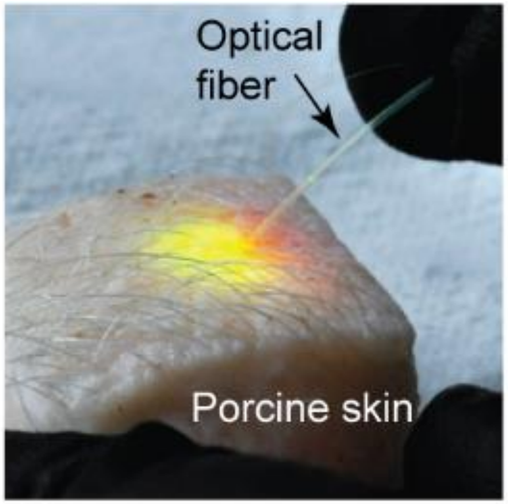


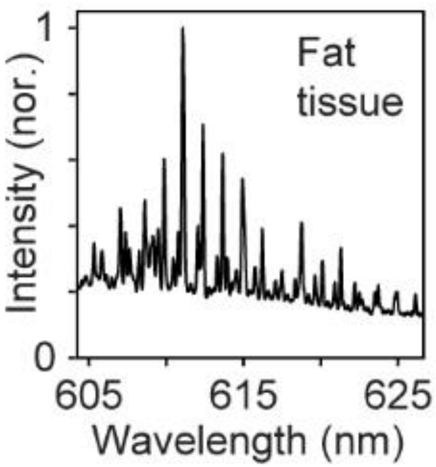


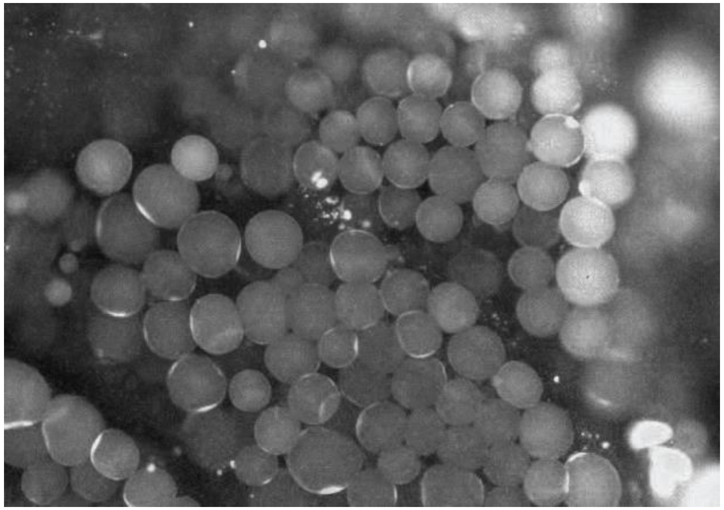


Lasing from subcutaneous fat tissue

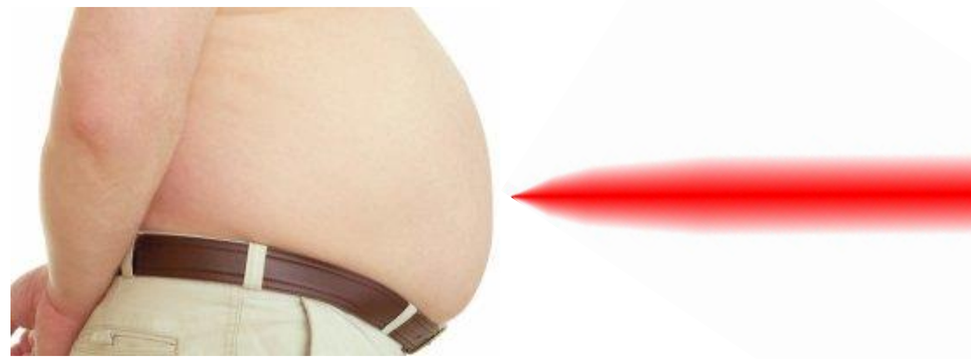
- Inject nile red dye
- Insert optical fiber
- Pump and collect laser light through the fiber



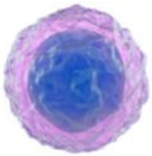




Rewriting science fiction



Media impact



Science
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nature photonics
nature methods
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the business of photonics
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Smithsonian
MIT Technology Review
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DiscoveryNews
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OSEBNOST PRIMORSKE

Osebnost Primorske meseca julija

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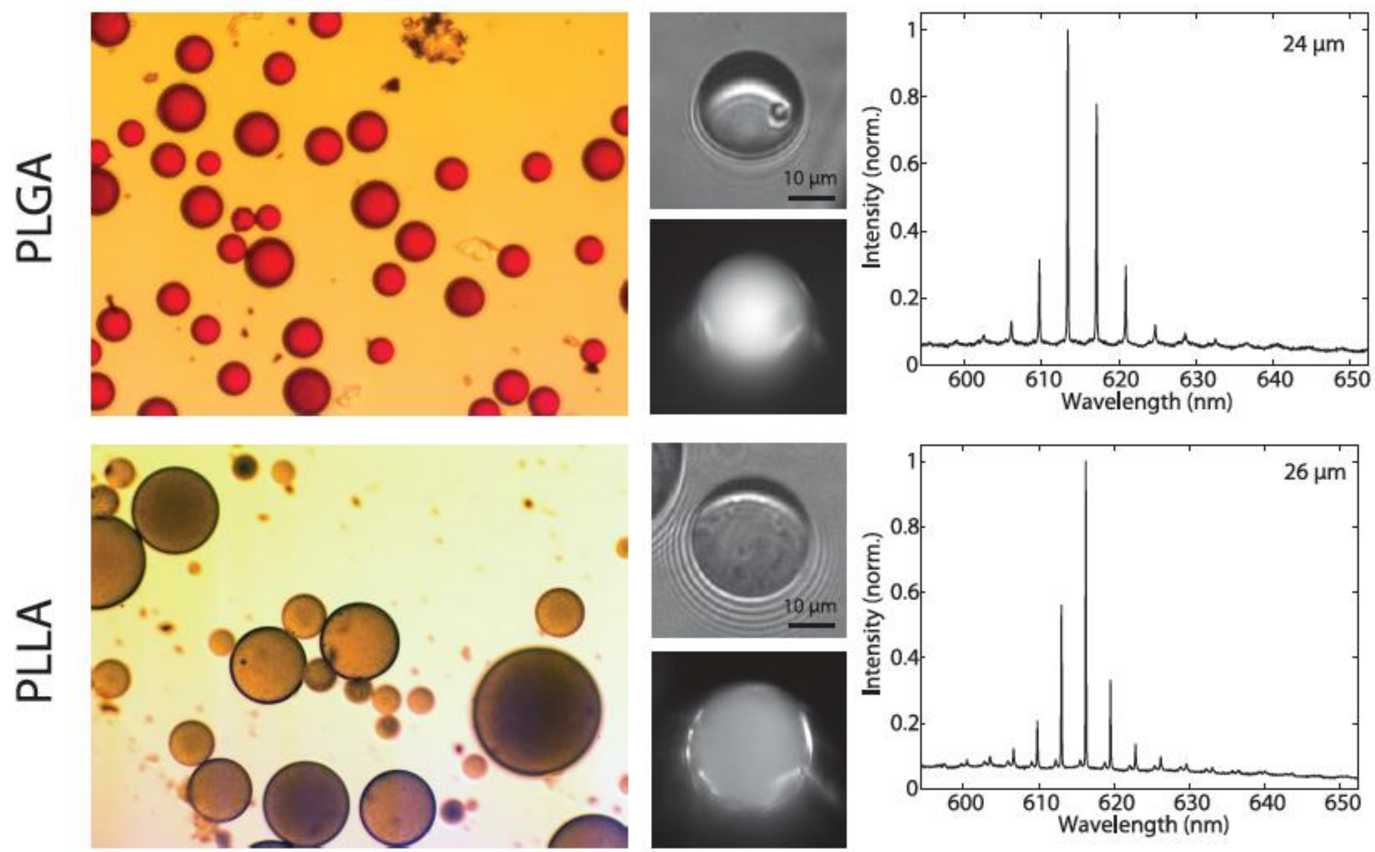
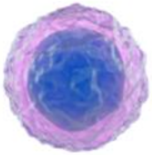
Biodegradable lasers

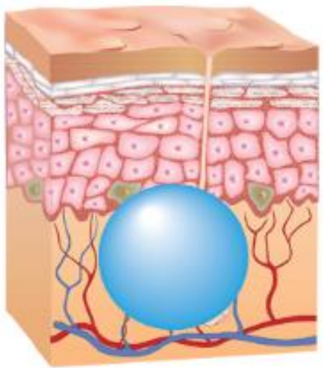
- Poly(lactic-co-glycolic acid) (PLGA) or Poly-L-lactide (PLLA)
- Dissolved in dichloromethane (DCM) + fluorescent dye
- Oil in water dispersion
- The minimum size for lasing: 20 µm

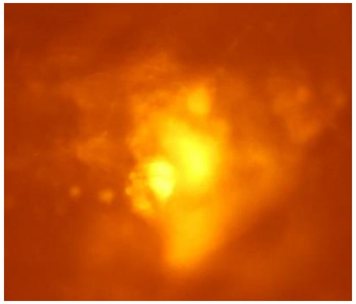


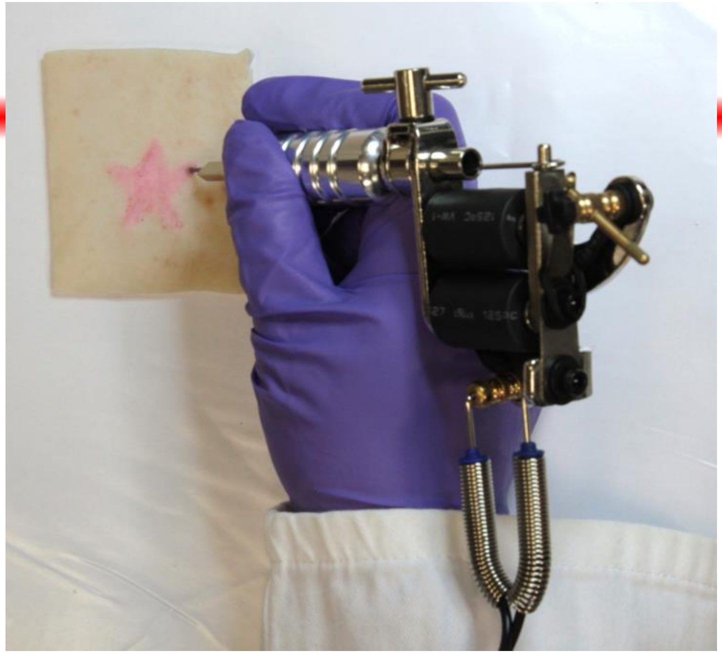


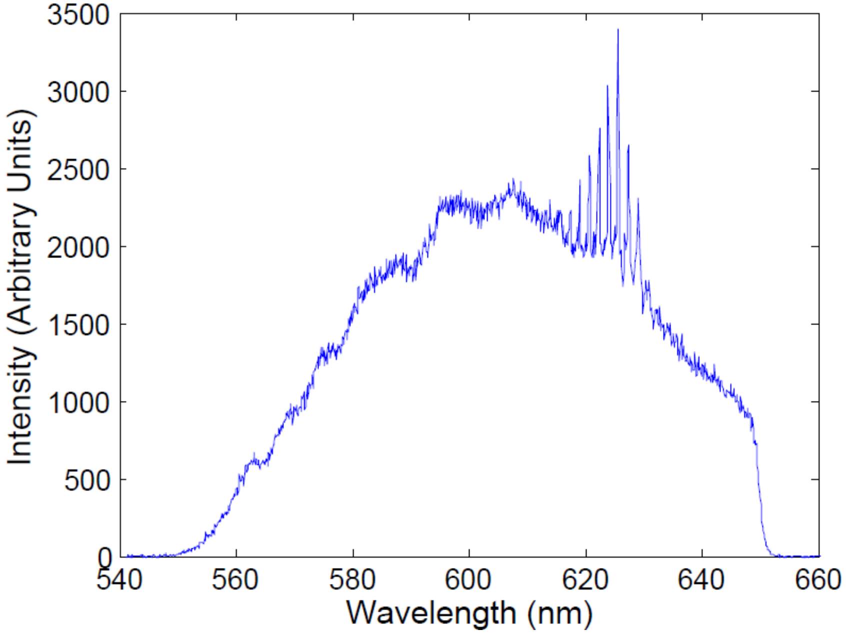
Laser tattoos

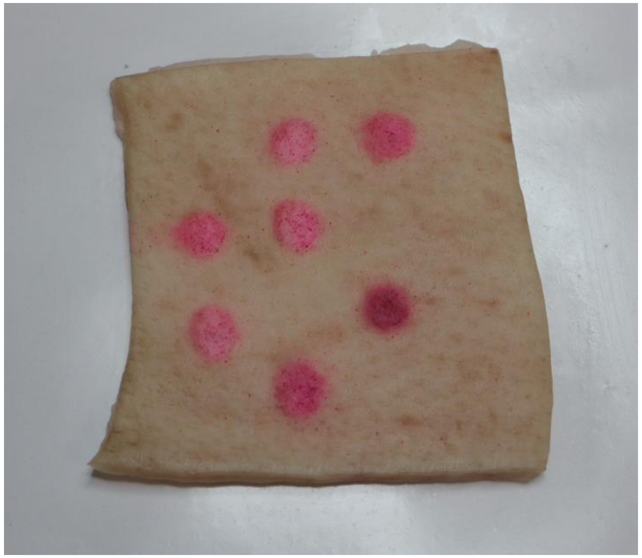
- Implantation of biocompatible lasers into skin using standard tattoo gun











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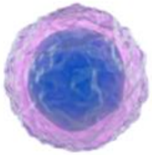
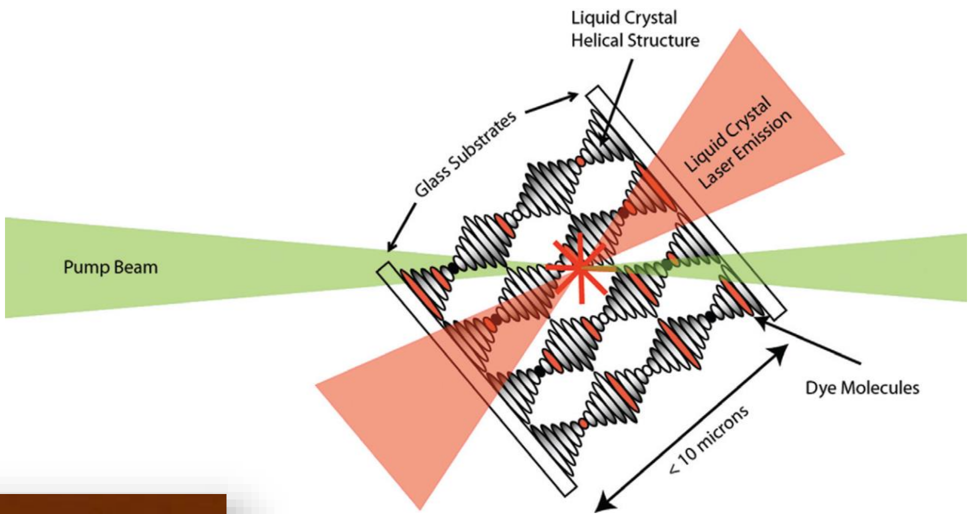
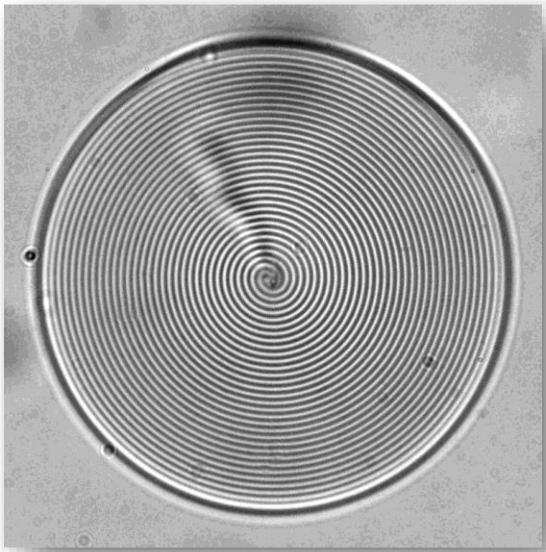


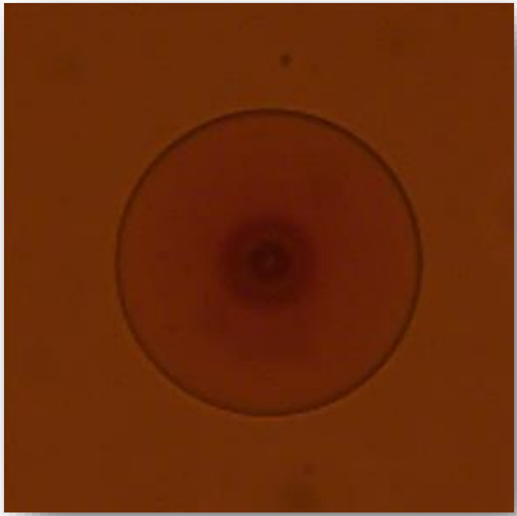
Cholesteric liquid crystal droplet laser

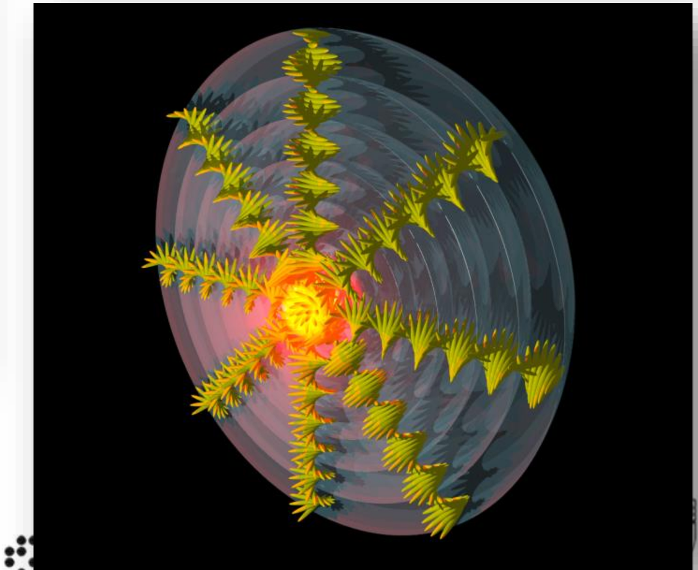
- Helical twist -> photonic crystal
- Dye doped LC droplet is a Bragg onion laser
- Single mode lasing
- Omnidirectional emission
- Smallest laser size: 14 µm



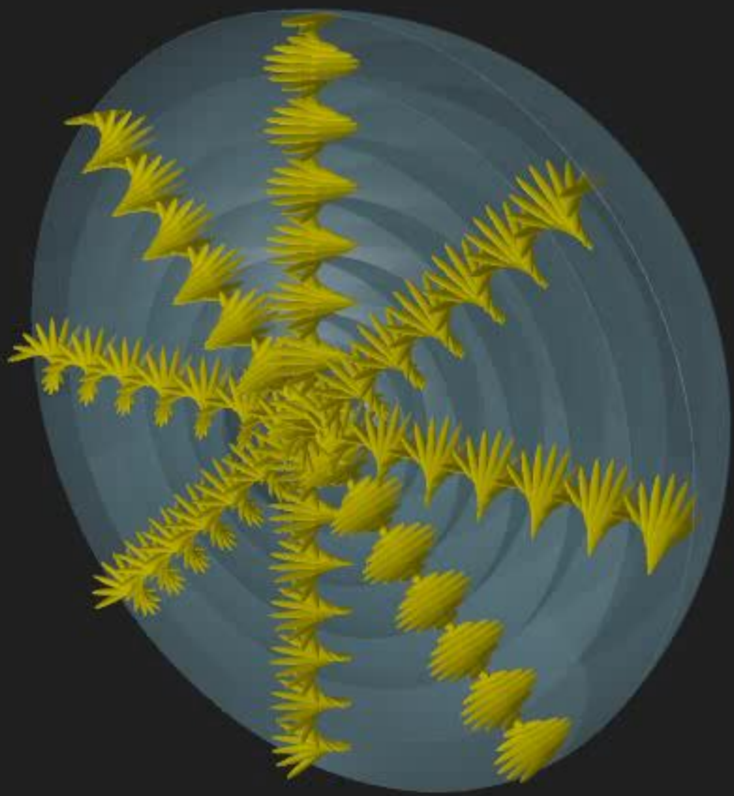
A. D. Ford, et al, Materials Today 9, 2006.





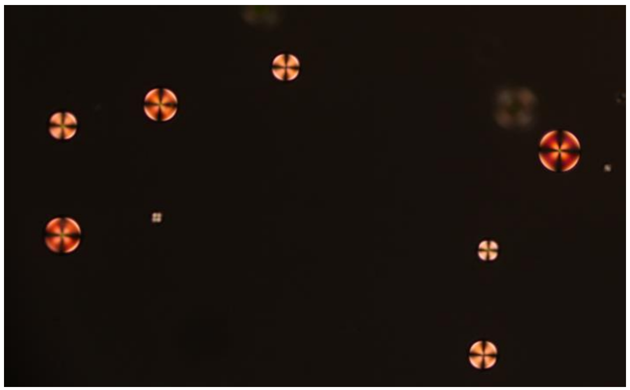


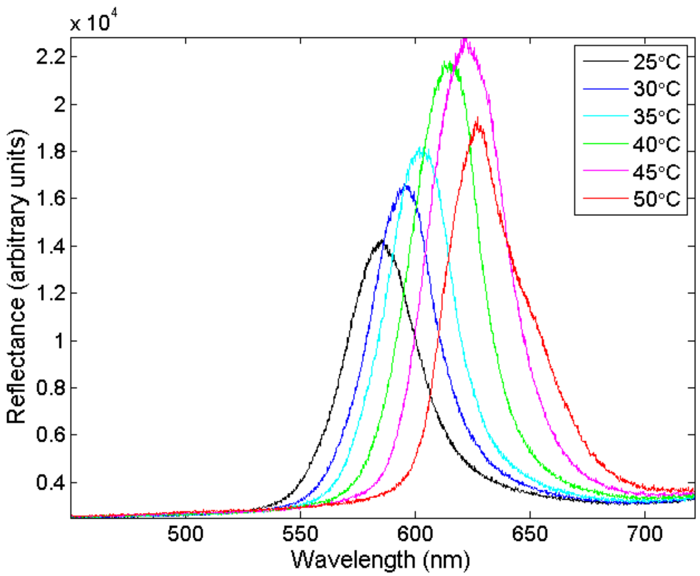
M. Humar and I. Muševič, 3D microlasers from self-assembled cholesteric liquid crystal microdroplets, *Opt. Express* **18**, 26995-27003 (2010).

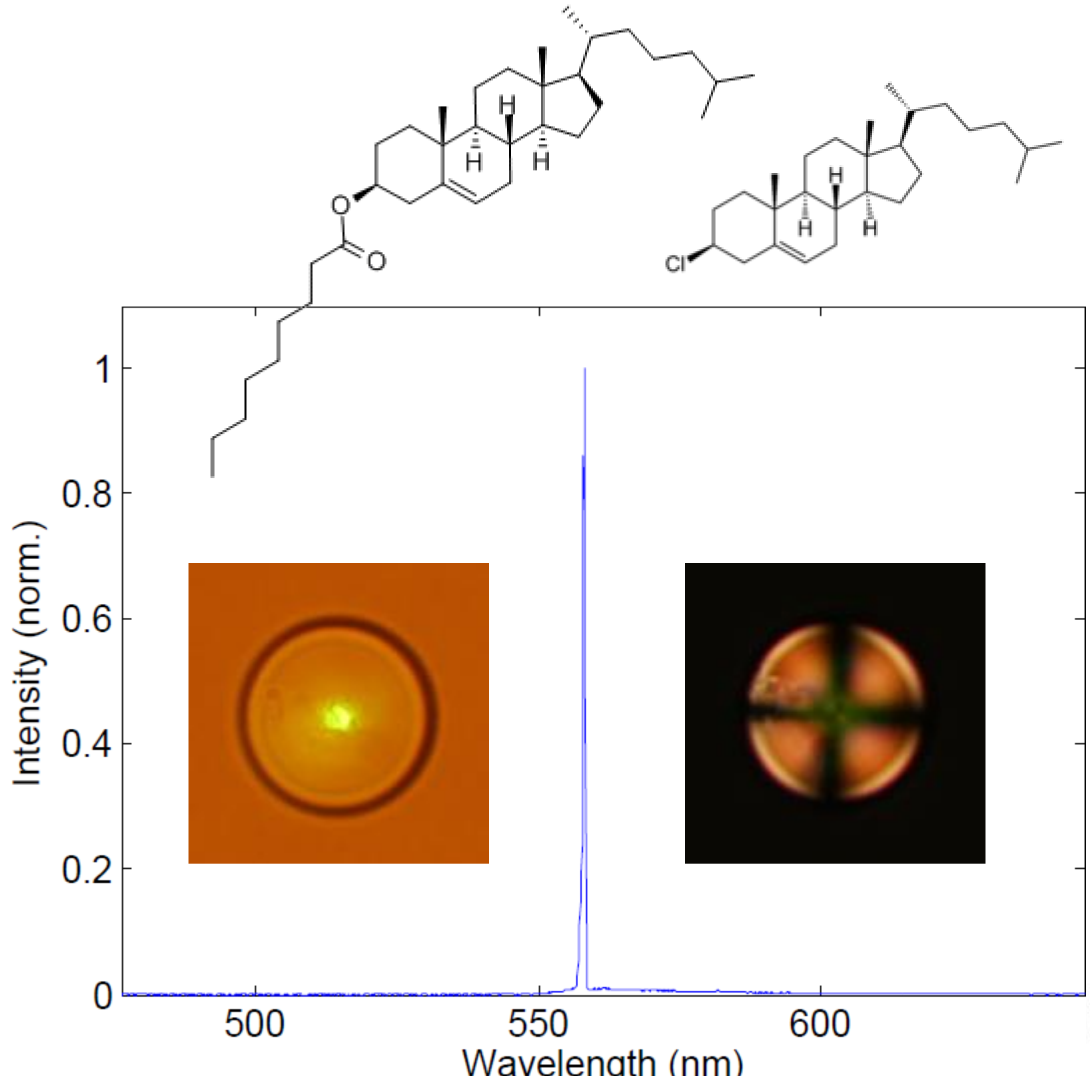


Cholesterol laser

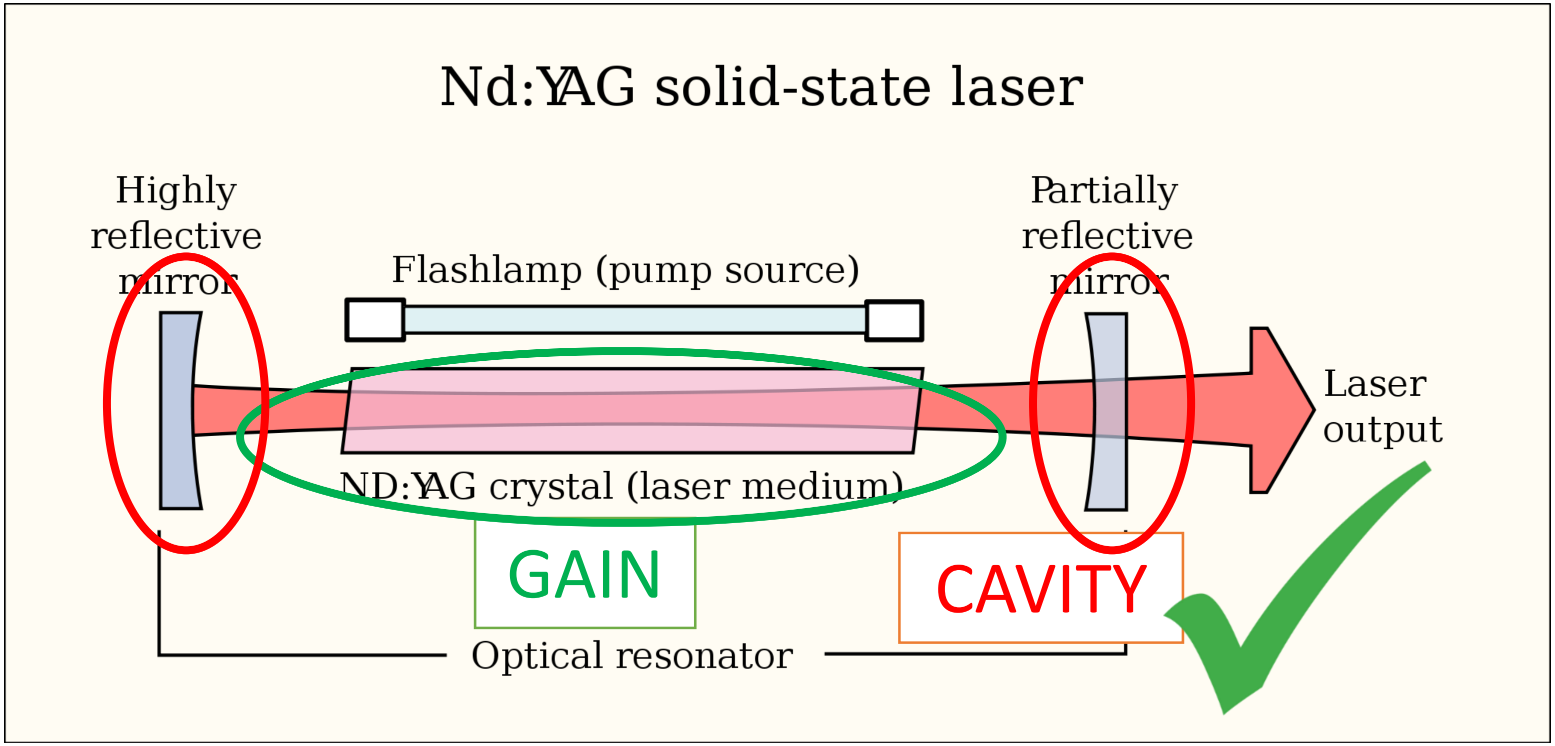
- Cholesteryl chloride, cholesteryl nonanoate and cholesteryl oleyl carbonate
- Biocompatible, since already present in our body







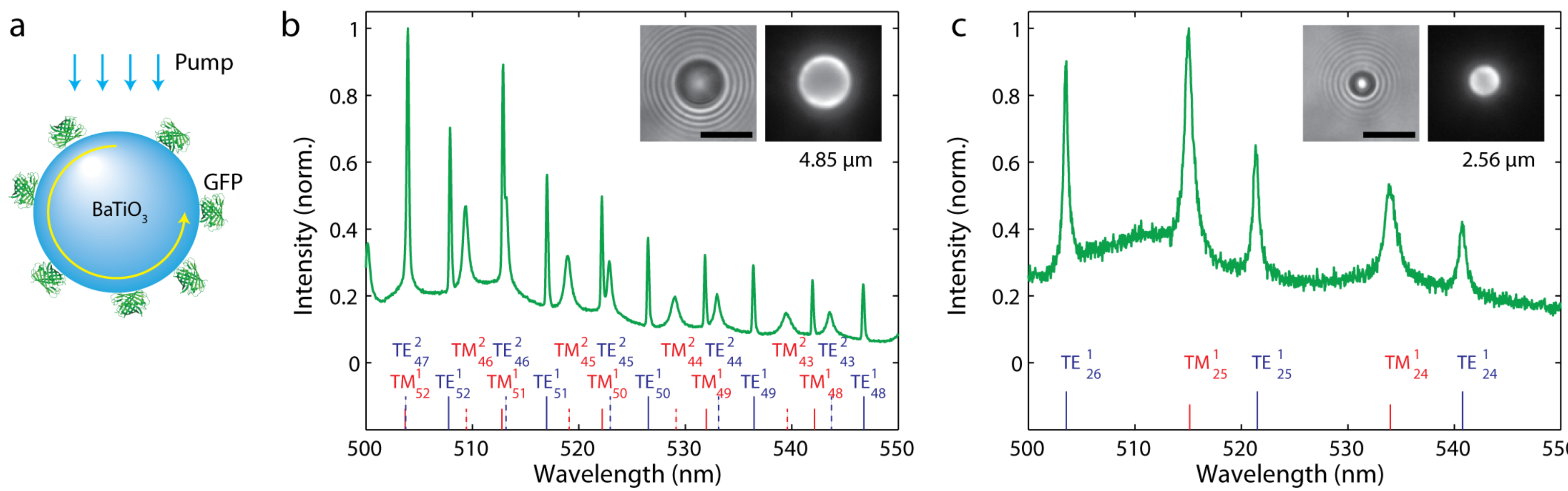
Lasers made completely out of biological materials

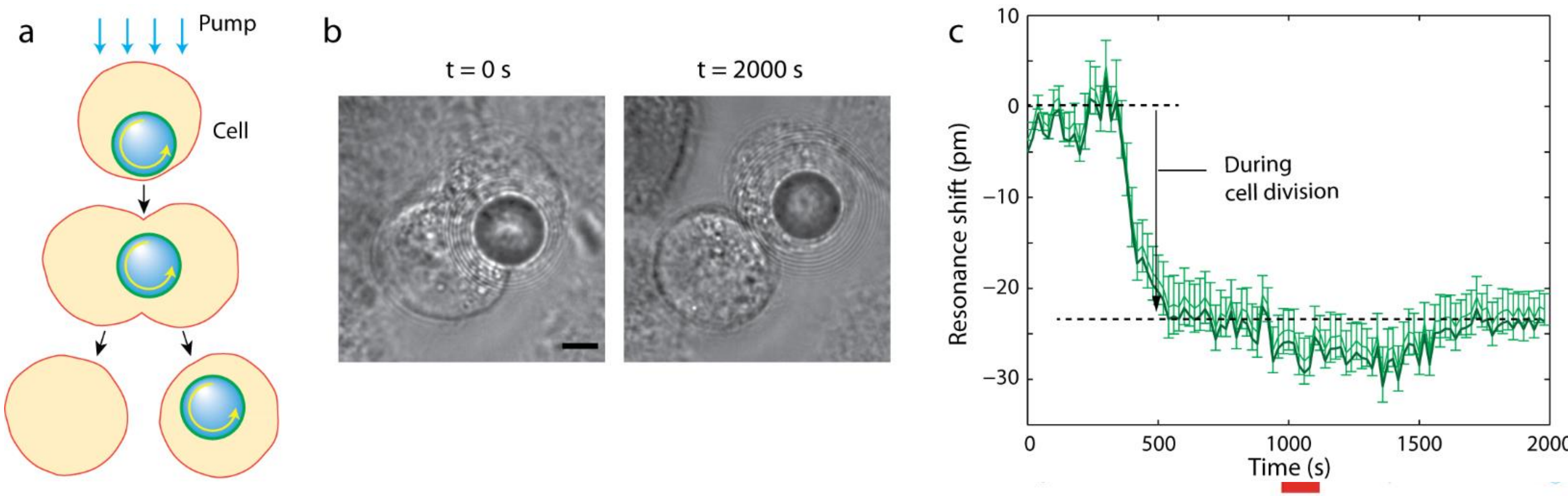


Wikipedia

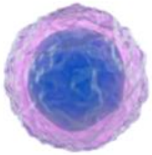
GFP WGM "lasers"

- GFP chemically linked to the surface of beads ($BaTiO_3$-APTES-GA-GFP)
- Whispering gallery modes generated

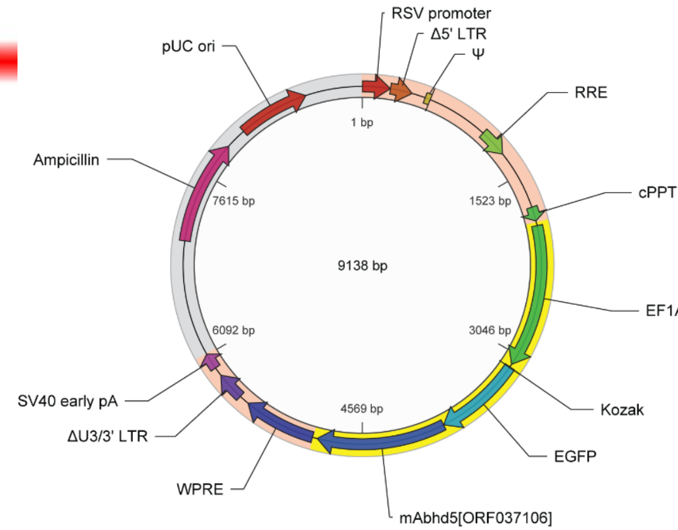


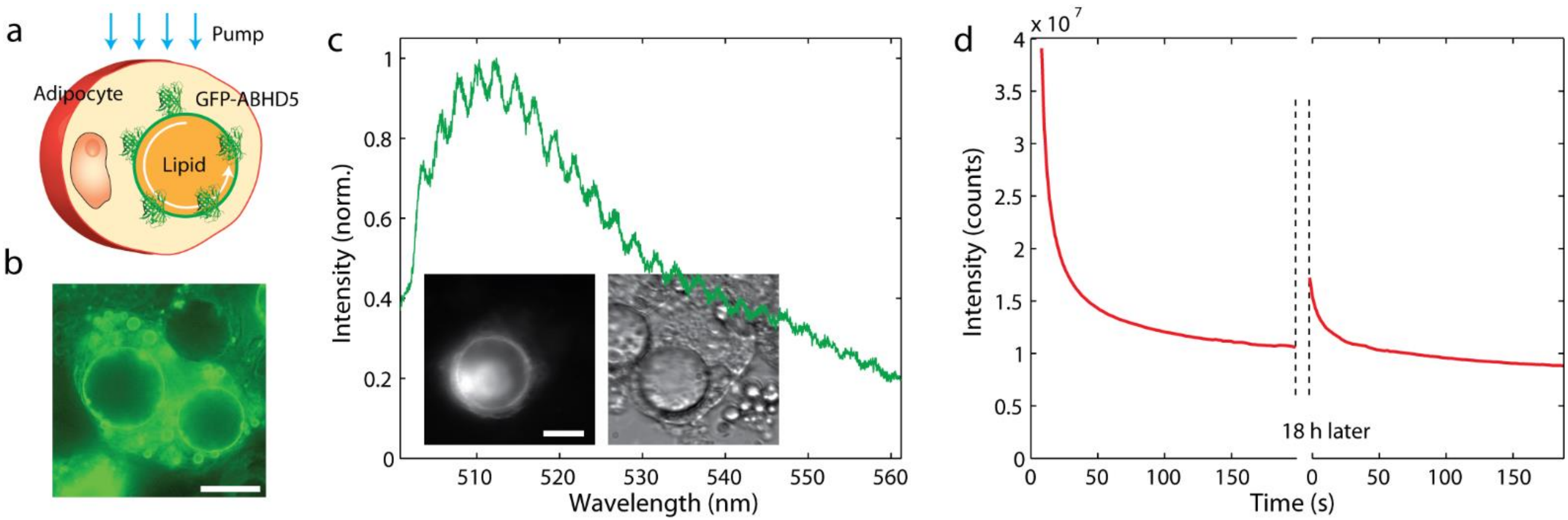


GFP in adopocytes



- Can we combine
  - Bio-cavity – lipid droplets in adipocytes
  - Bio-gain – GFP transfection
- We designed a custom fusion protein: Abhd5-GFP
- Abhd5 is known to bind to the surface of lipid droplets
- Completely bio-cavity, self-healing





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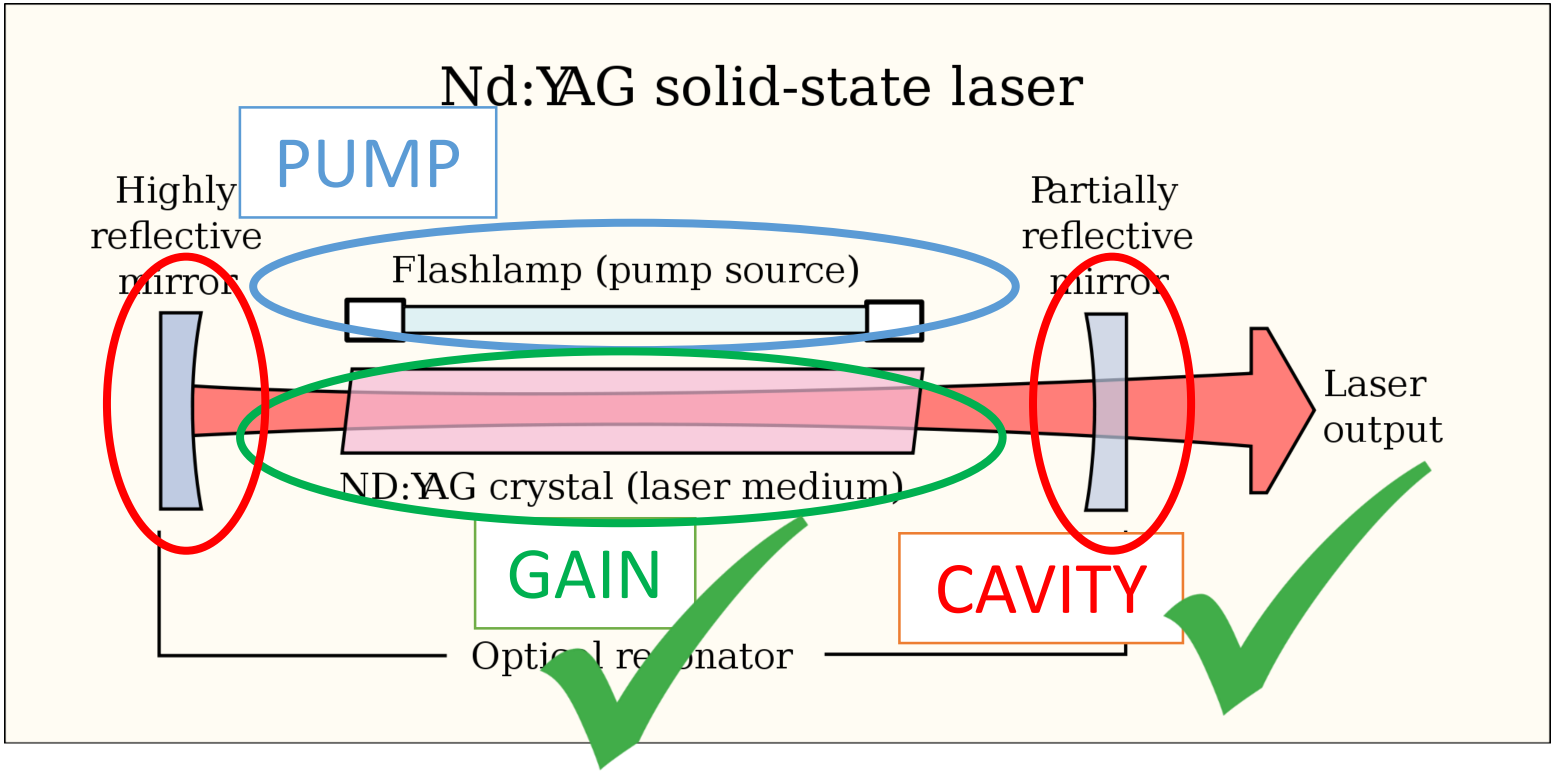
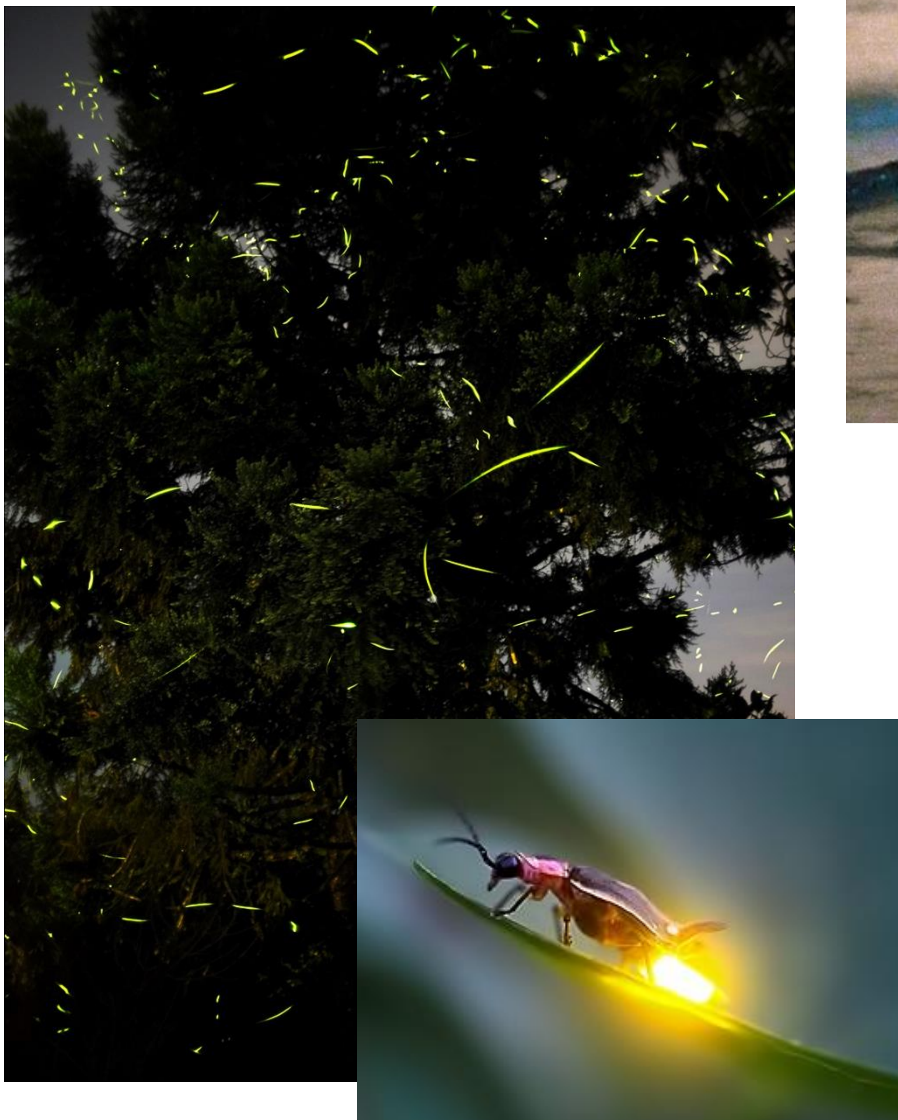
Lasers made completely out of biological materials

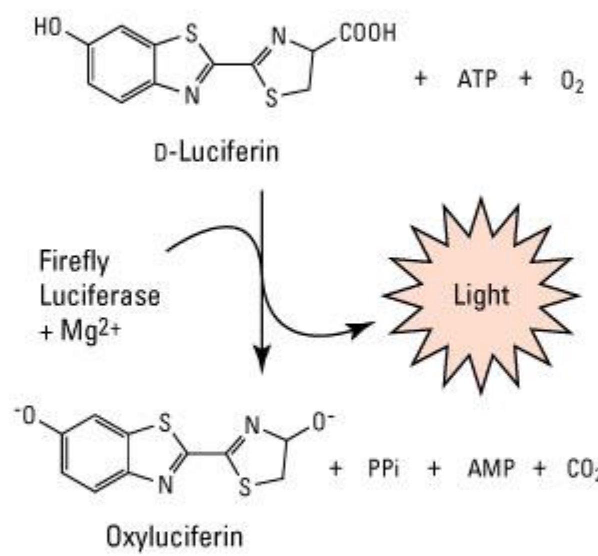


Wikipedia

Bioluminiscence

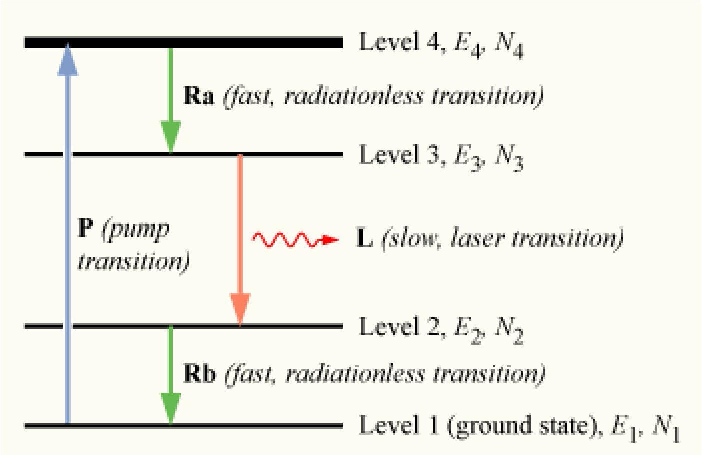


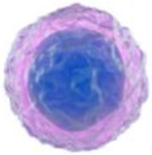
luciferaze + luciferin -> light



Pumping a laser with bioluminescence

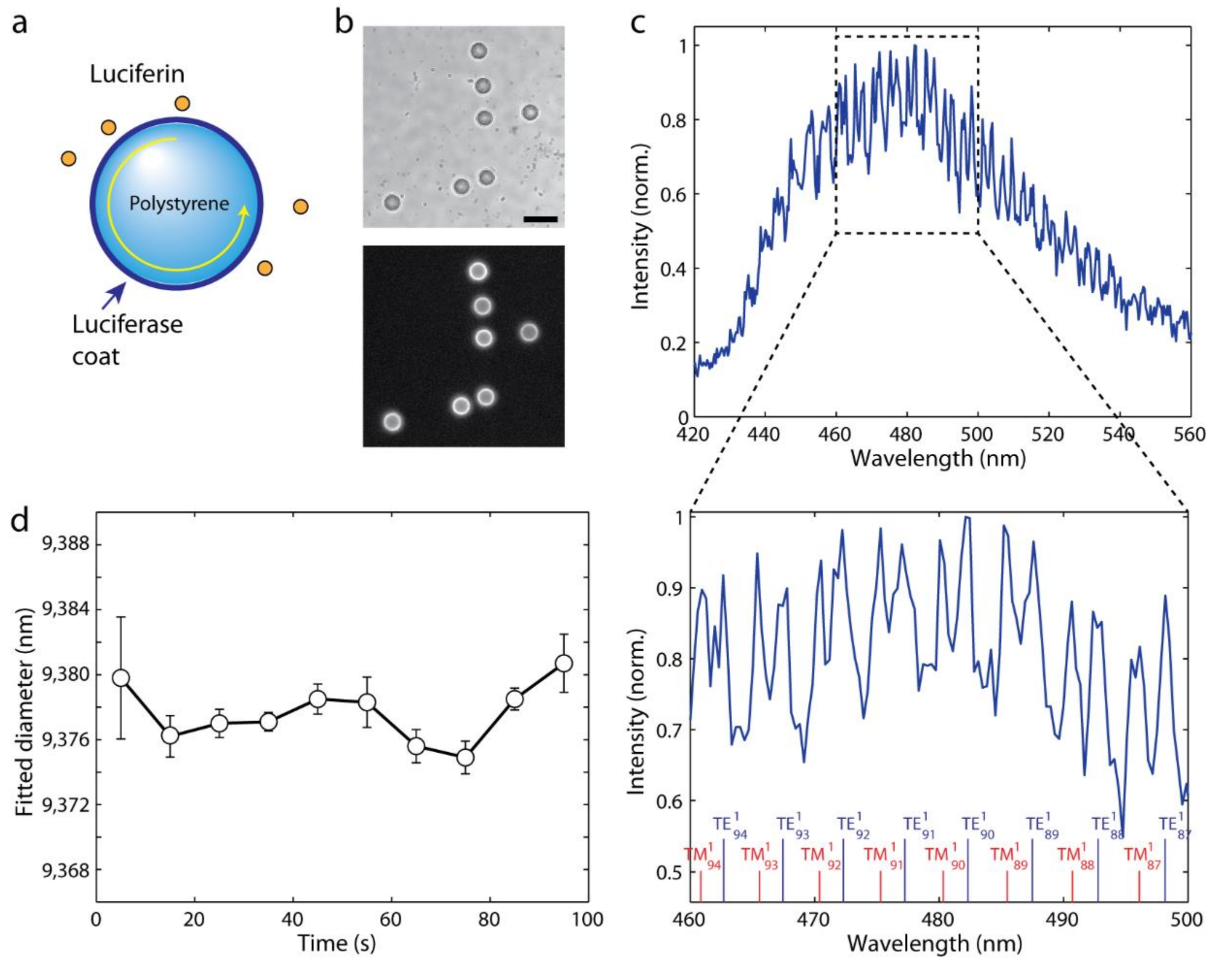
- Bioluminescence to create population inversion
- Rate of photon generation: ~ 0.2 photons/molecule/s
- Excited states lifetime: few nanoseconds
- Ten orders of magnitude difference
- Chemical lasers only demonstrated using highly reactive gases and reacting them at supersonic flows





Bioluminescence pumped WGMs below threshold

- Beads coated with luciferase
- Introducing luciferin into solution
- Light generated is coupled to WGMs
- Laser like emission



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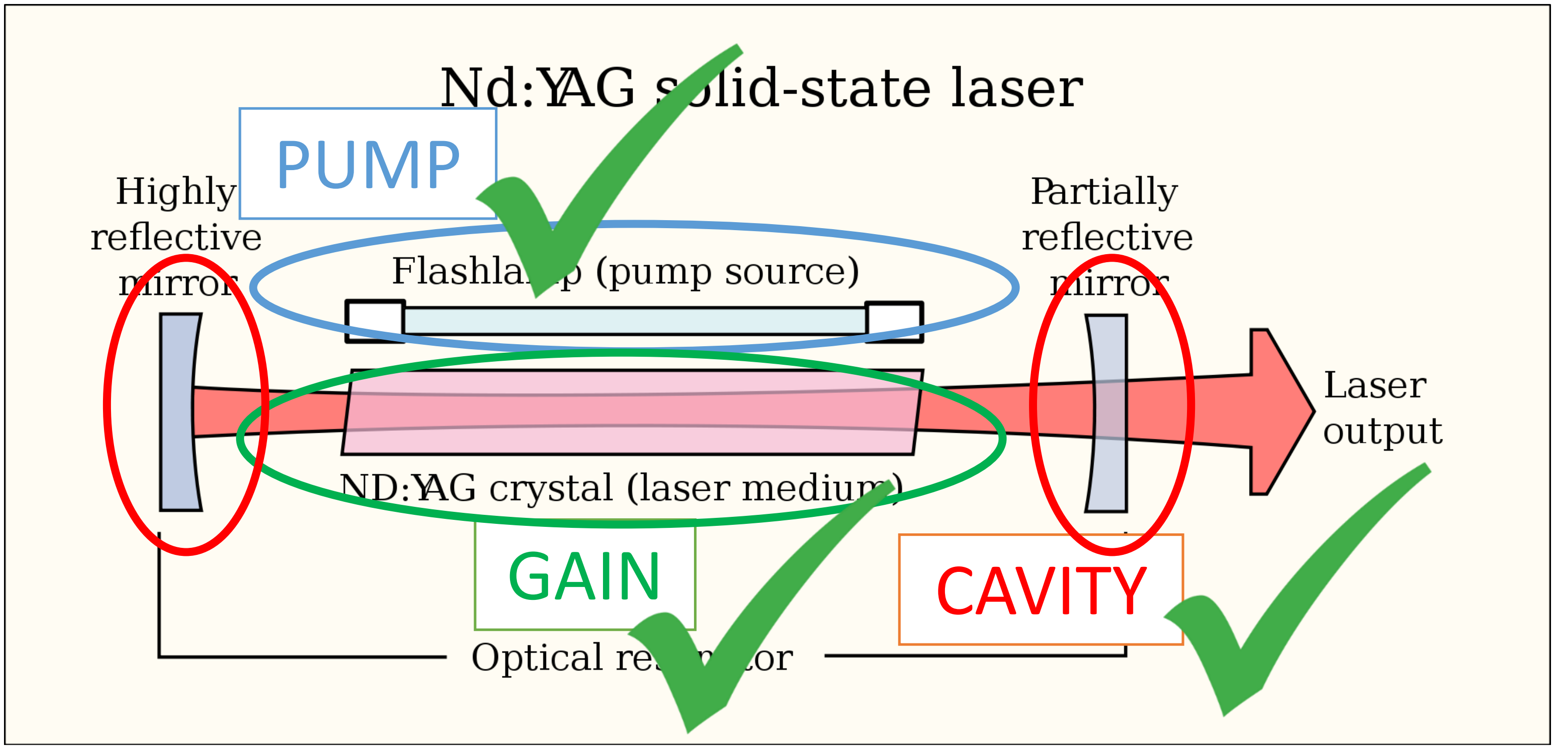
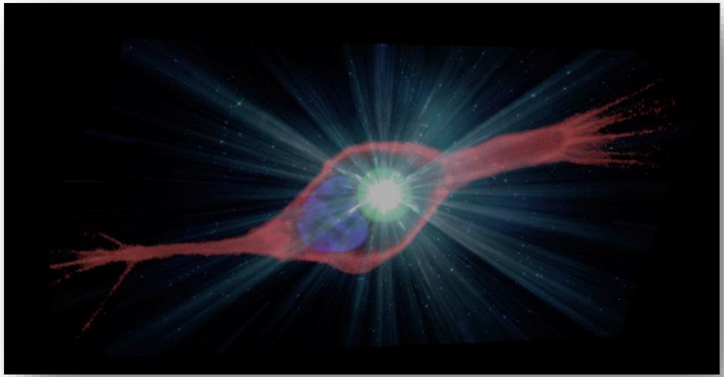
Lasers made completely out of biological materials



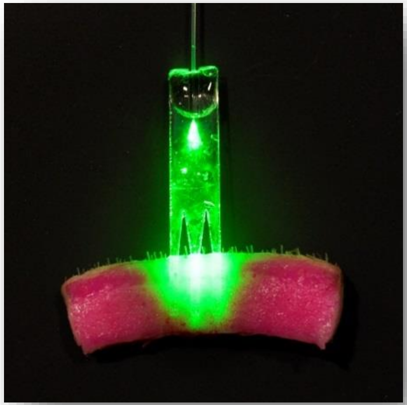
Wikipedia

Contents

- Cell lasers



- Biocompatible optical waveguides



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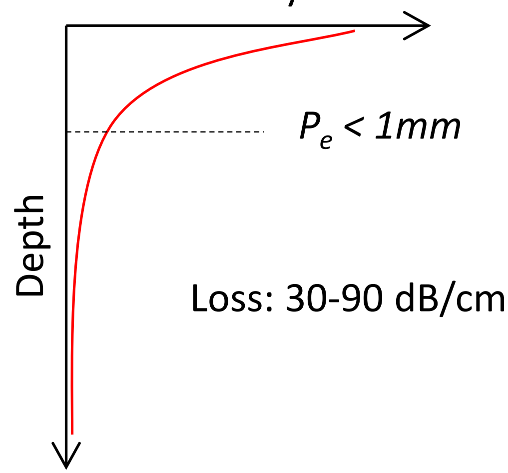
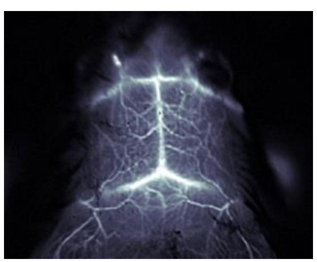
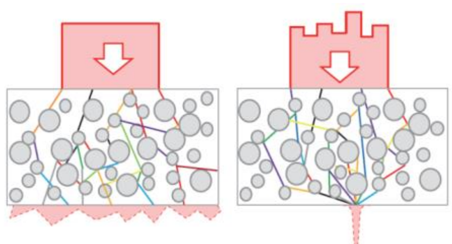
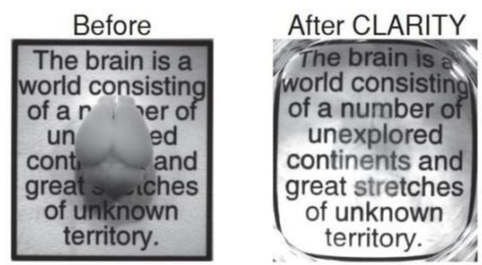
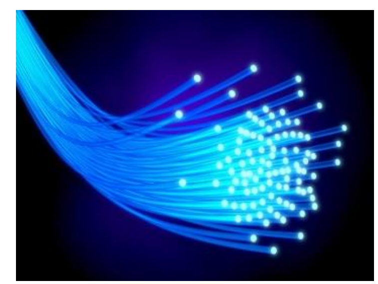
Light penetration depth

Tissue



NIR window	Wavefront shaping	Tissue clearing	**Waveguide**
			
Nat Photon, 2014	*Nat Photon, 2013*	*Nature 2013*	

Limited penetration

ex vivo

Non-biocompatible

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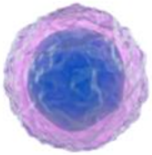
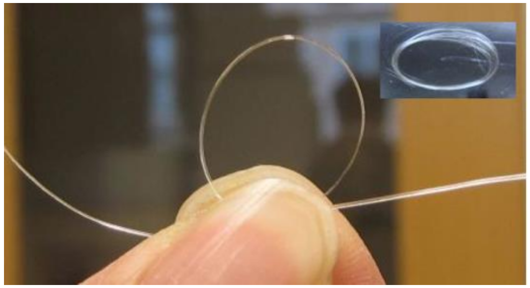
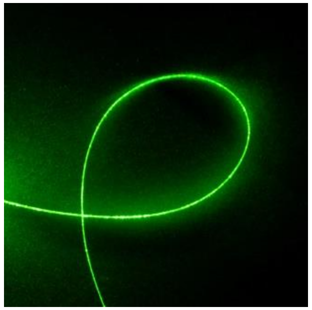


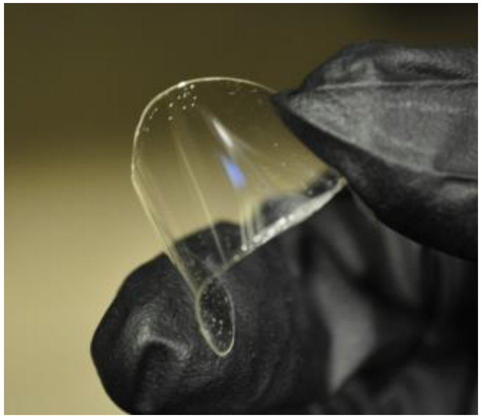
Biomaterial Optical Fiber





- Materials:
  - Poly lactic co-glycolic acid (PLGA)
  - Polylactic acid (PLA)
- Properties
  - Routinely used for medical purposes
  - Biodegradable by hydrolysis
- Fabrication
  - Melting and drawing
  - Laser cutting

→



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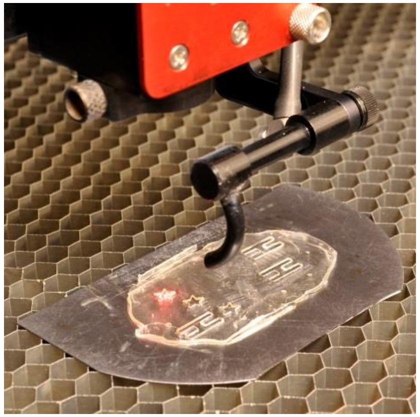
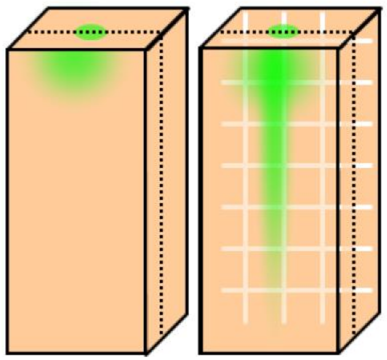
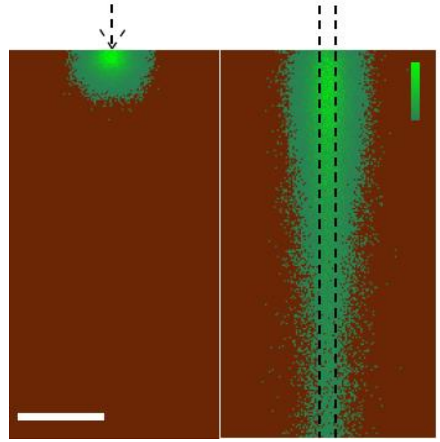


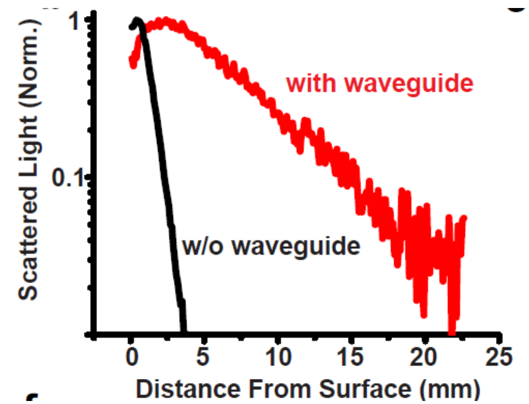
Nizamoglu, Gather, **Humar**, et al., Nature Communications 2015.

A comparative study of light penetration

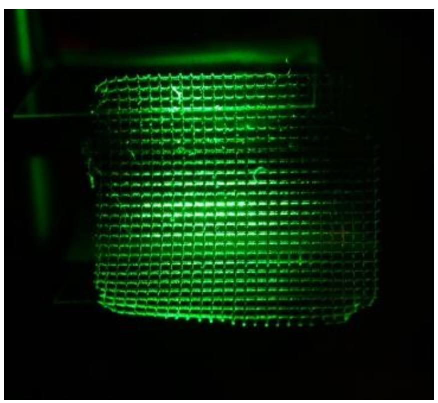
Monte Carlo Simulation

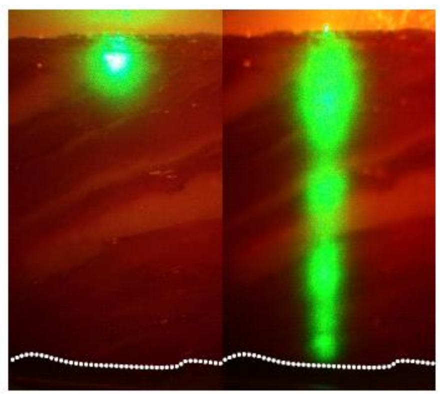


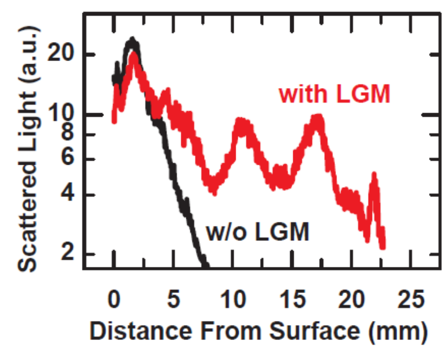




Experiment







Nizamoglu, Gather, **Humar**, et al., Nature Communications 2015.

Photochemical tissue bonding (PTB)

- Procedure
  - Apply dye to wound & Irradiate
    - Dye: Rose Bengal (FDA-approved) & Green light
  - Covalent crosslinks between amino acids in collagen molecules
- Properties of PTB
  - Fast
  - Water-tight seal
  - Non-inflammatory
  - Minimal scarring

Lasers Demonstrate the Power to Heal Without Scarring

Green laser light can trigger collagen fibers to link up in nerves and other damaged tissue

By Larry Greenemeier

Scientific American, 50th anniversary of laser (2010)

<u>Skin depth problem (1 mm @1 W/cm^2)</u>

Yang P et al. Lasers Surg. Med. 44:163 (2012).
S. Tsao et al. British Journal of Dermatology 166, 555 (2012)
T.S. Johnson et al. J. Surg. Res. 143, 224 (2007).
B. P. Chan, I. E. Kochevar, R. W. Redmond. J. Surg. Res. 108:77-84 (2002).

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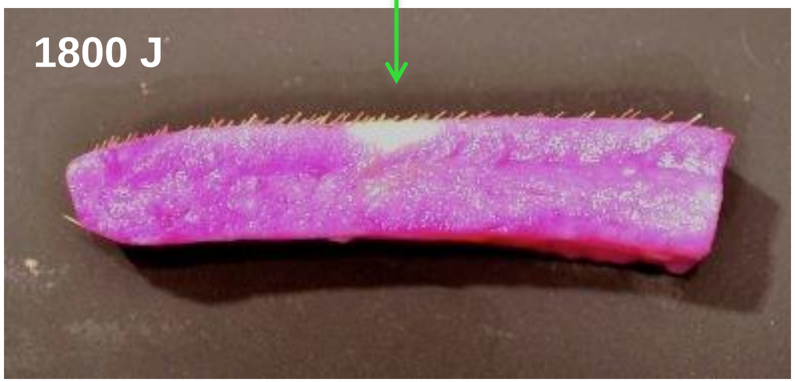
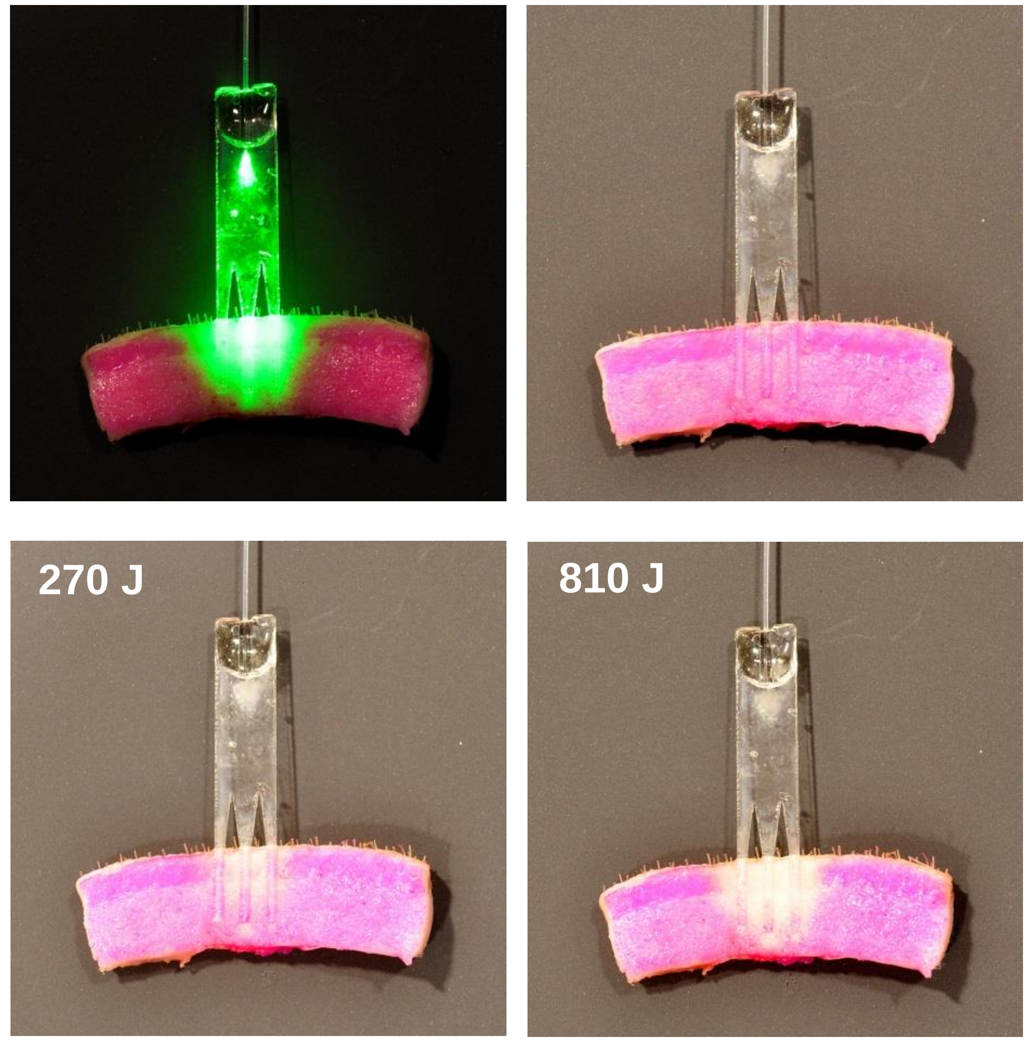
Waveguide assisted light penetration

Without waveguide



Waveguide assisted



Nizamoglu, Gather, **Humar**, et al., Nature Communications 2015.

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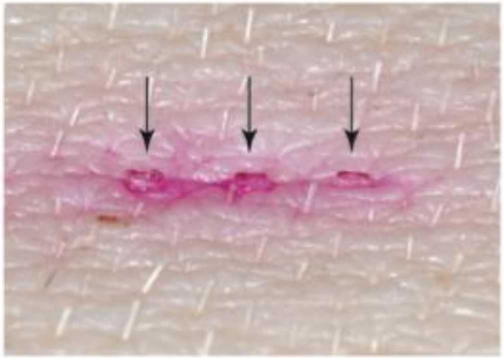
Waveguide assisted PTB procedure



Nizamoglu, Gather, **Humar**, et al., Nature Communications 2015.

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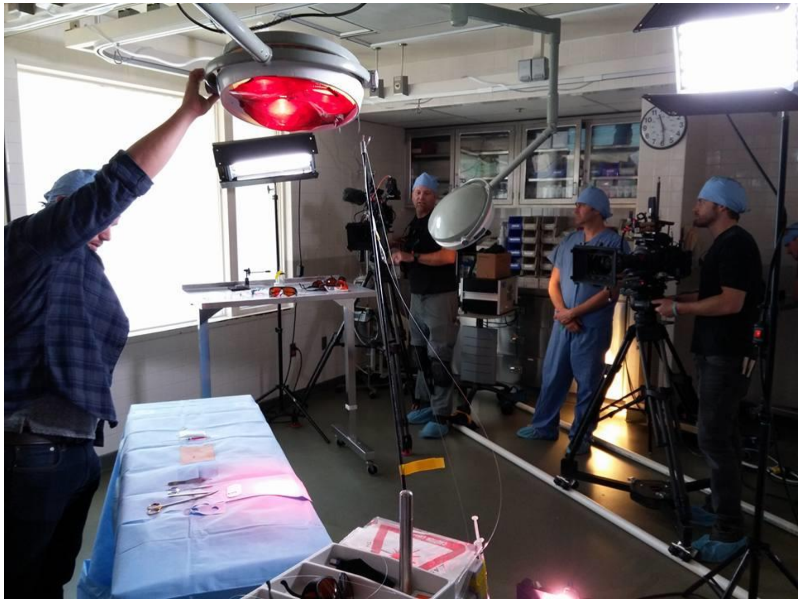
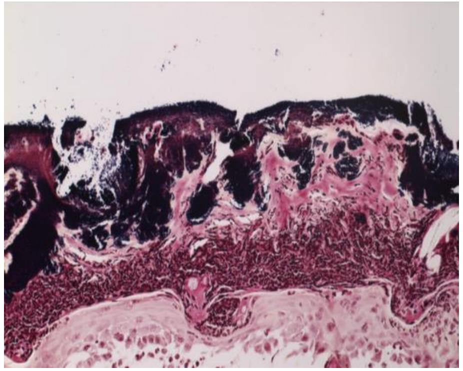
The White Rabbit Project



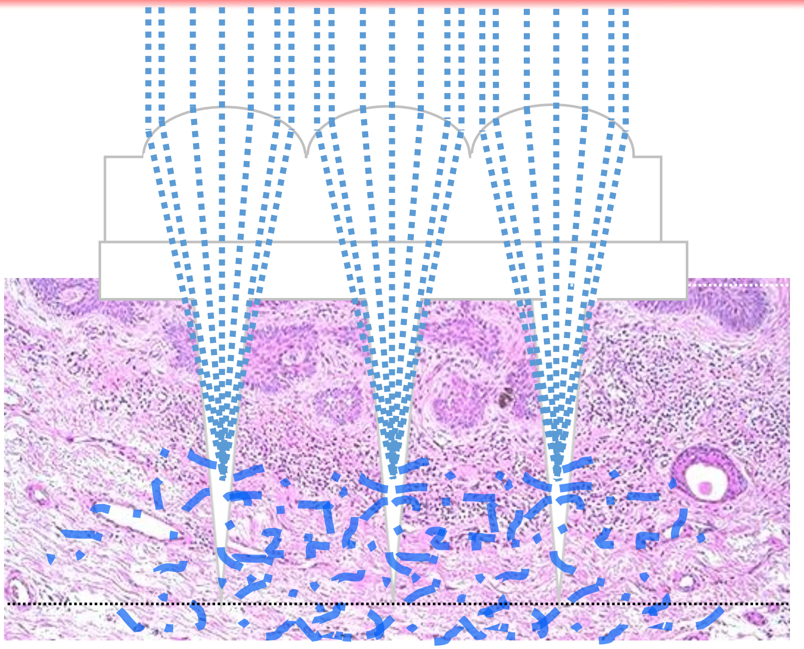
Tory Belleci from Mythbusters

Delivery of blue light into skin

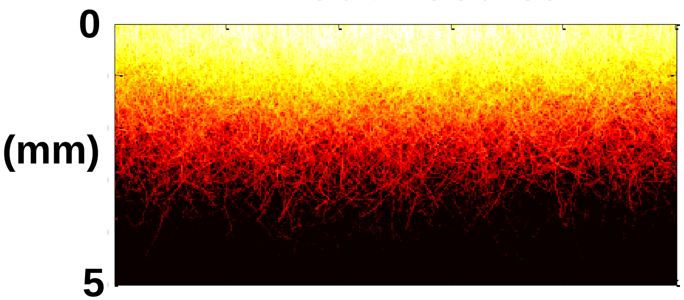
- Treatment of skin conditions
- Example: blue light bacteria killing



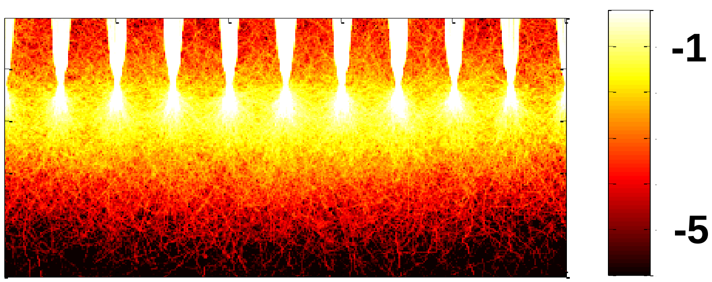
Methicillin-resistant Staphylococcus aureus (MRSA) infection



Without needles



With needles



Wavelength : 491 nm

Color bar : intensity in log scale

M Kim, J An, KS Kim, M Choi, **M Humar**, SJJ Kwok, T Dai, SH Yun, Optical lens-microneedle array for percutaneous light delivery, Biomedical Optics Express 7, 4220-4227 (2016).

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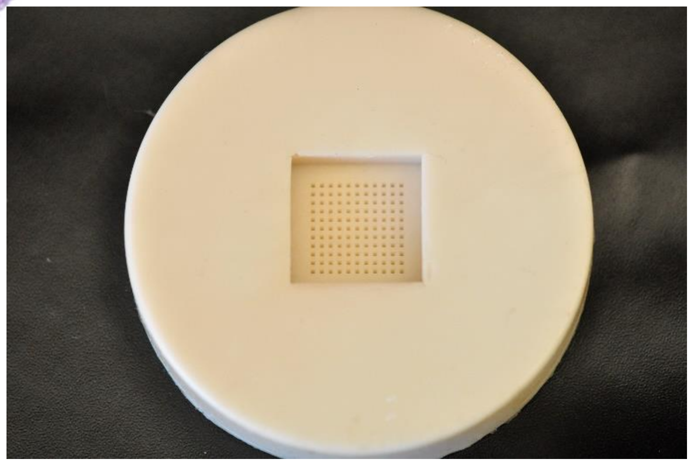
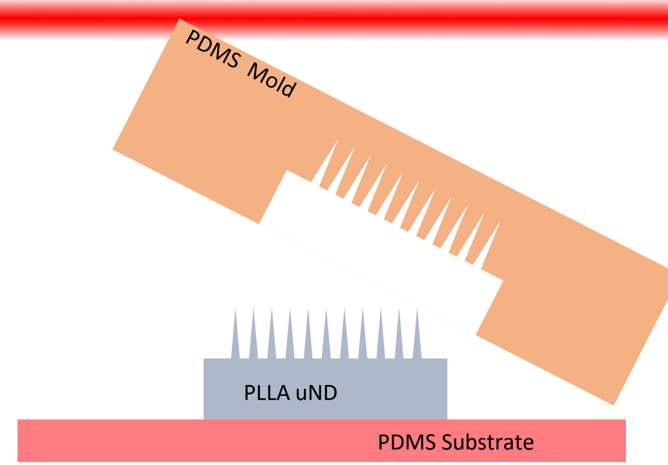
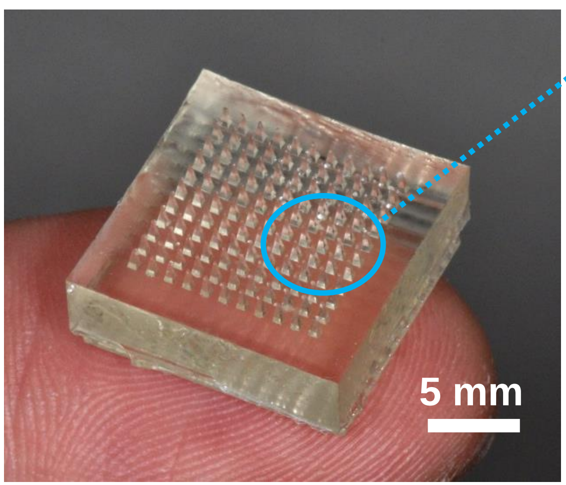
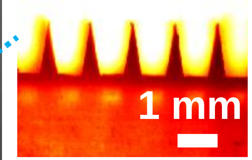
Fabrication of Lenslet-MNA



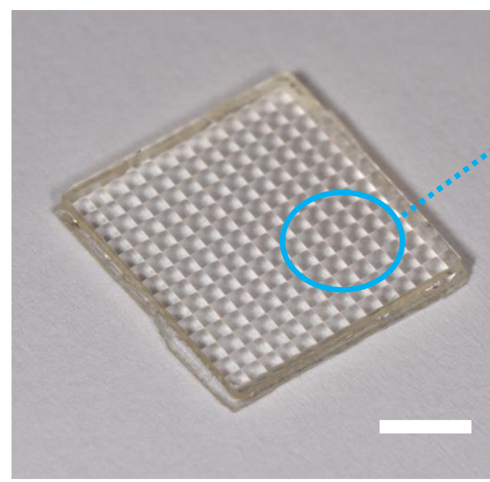
MNA Mold

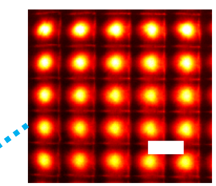






Length : 1600 µm
Interspacing : 1mm
Area: 10 mm X 10 mm





Focal Length : 9.5 mm
Interspacing : 1mm

M Kim, J An, KS Kim, M Choi, **M Humar**, SJJ Kwok, T Dai, SH Yun, Optical lens-microneedle array for percutaneous light delivery, Biomedical Optics Express 7, 4220-4227 (2016).

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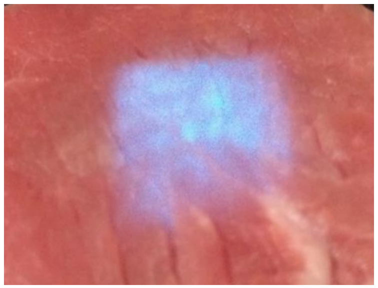
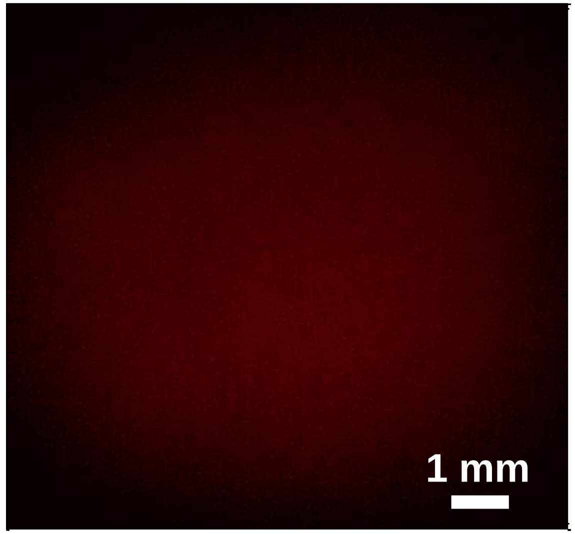
Enhanced Transmission through tissue

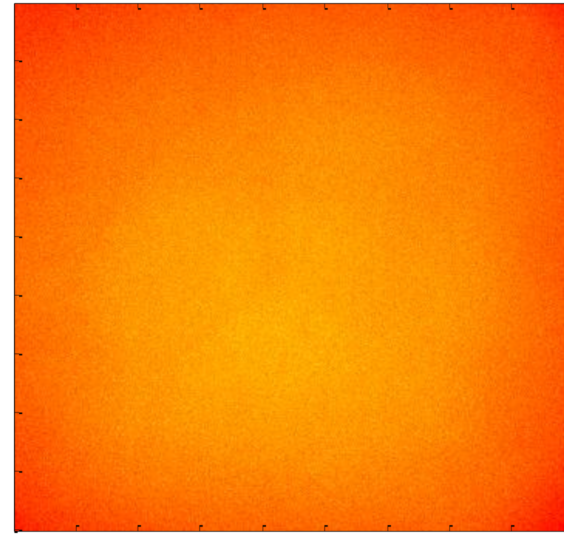
	W/O OMN	W/ OMN
Bovine 3.1 mm		



Enhancement factor: 9

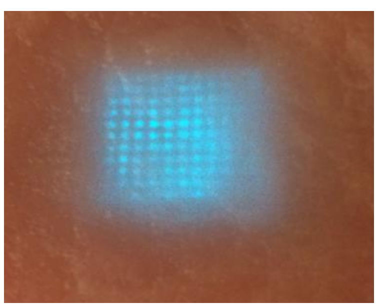


Transmittance: 0.85 %

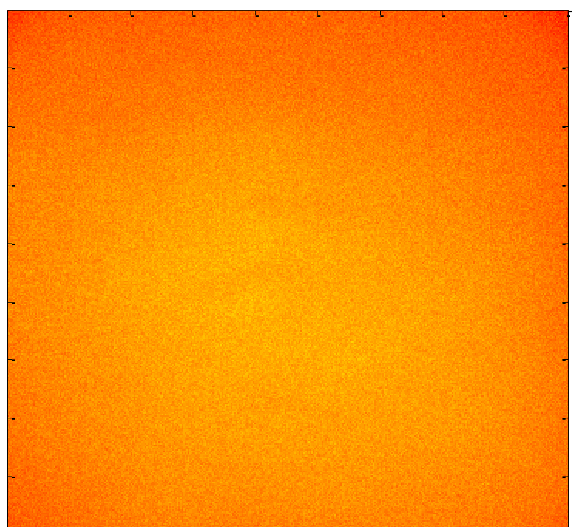


Transmittance: 7.6 %

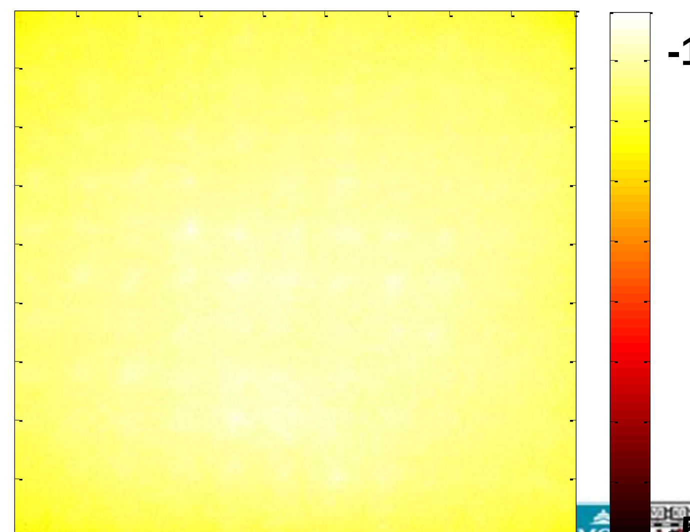
Porcine 2.7 mm



Enhancement factor: 4

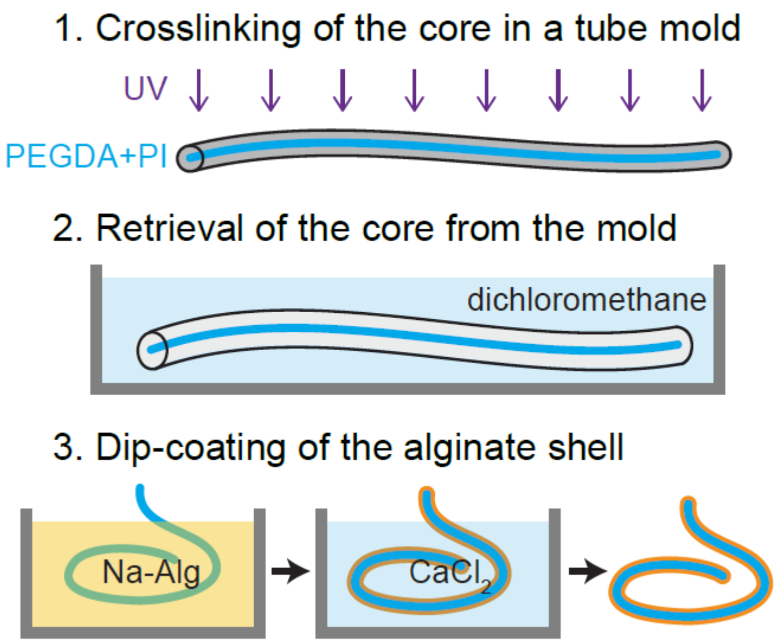


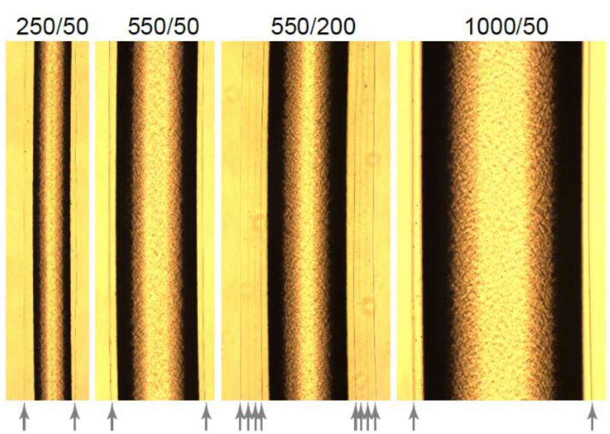
Transmittance: 8.3 %

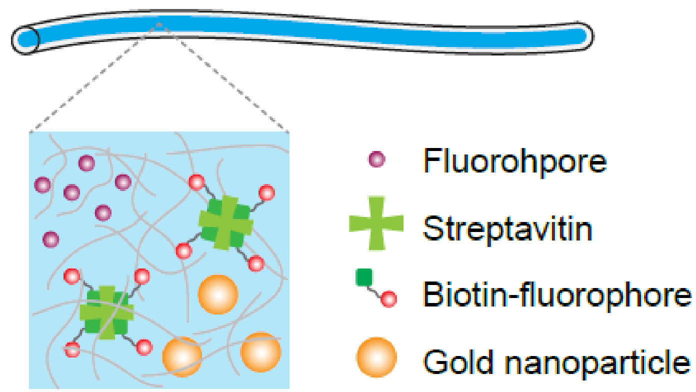


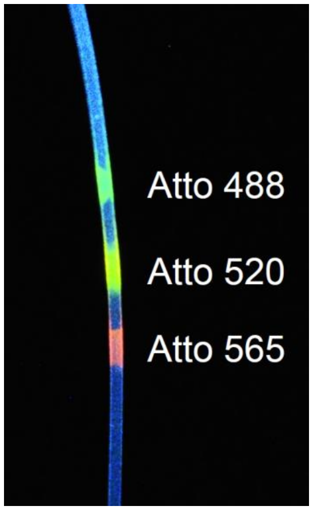
Transmittance: 32 %

Core-shell hydrogel optical waveguides









Choi, **Humar**, Kim, Yun, Advanced Materials 2015

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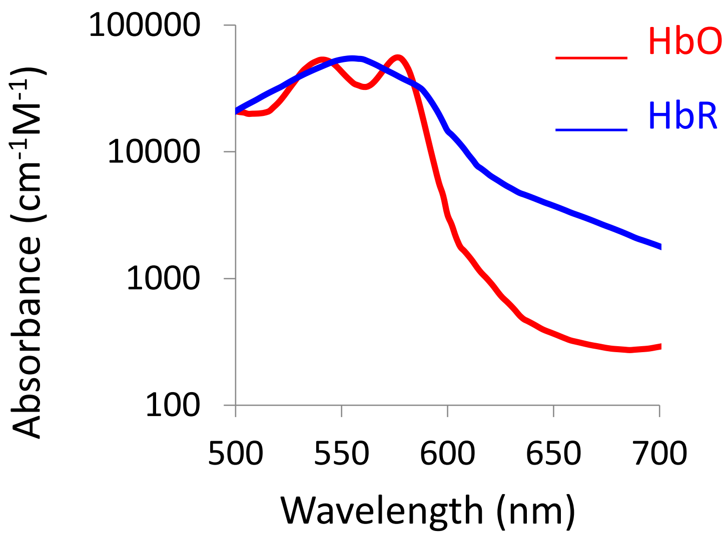
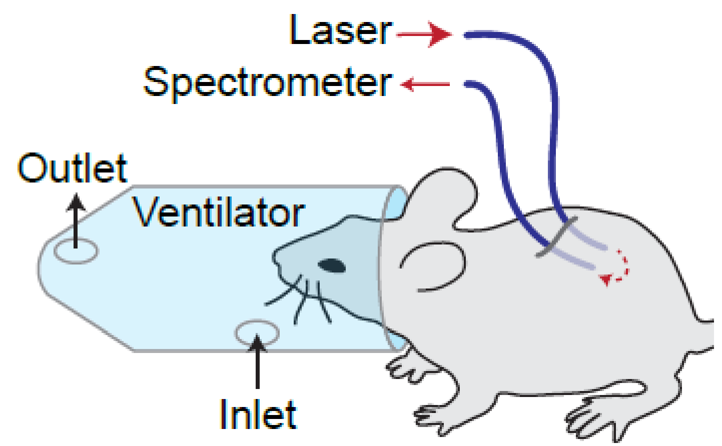
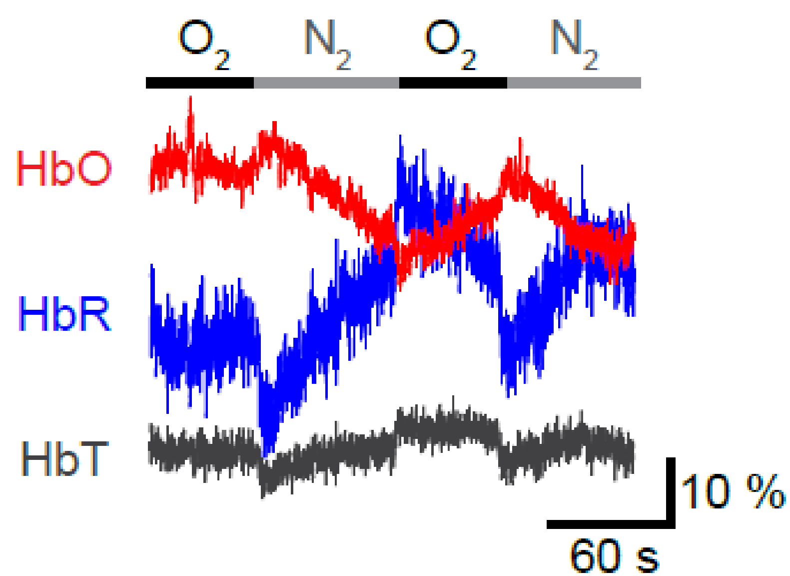
In vivo oxygen sensing







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Acknowledgements

HARVARD
Seok-Hyun (Andy) Yun
Mark Choi
Avinash Upadhya
Giuliano Scarcelli
Sedat Nizamoglu
Moonseok Kim
Ki Su Kim
Heebeom Koo
Malte Gather

JOŽEF STEFAN INSTITUTE
Anja Dobravec
Igor Muševič

Grants
Marie Curie International Outgoing Fellowship
Marie Curie Reintegration Fellowship
Direktorjev sklad

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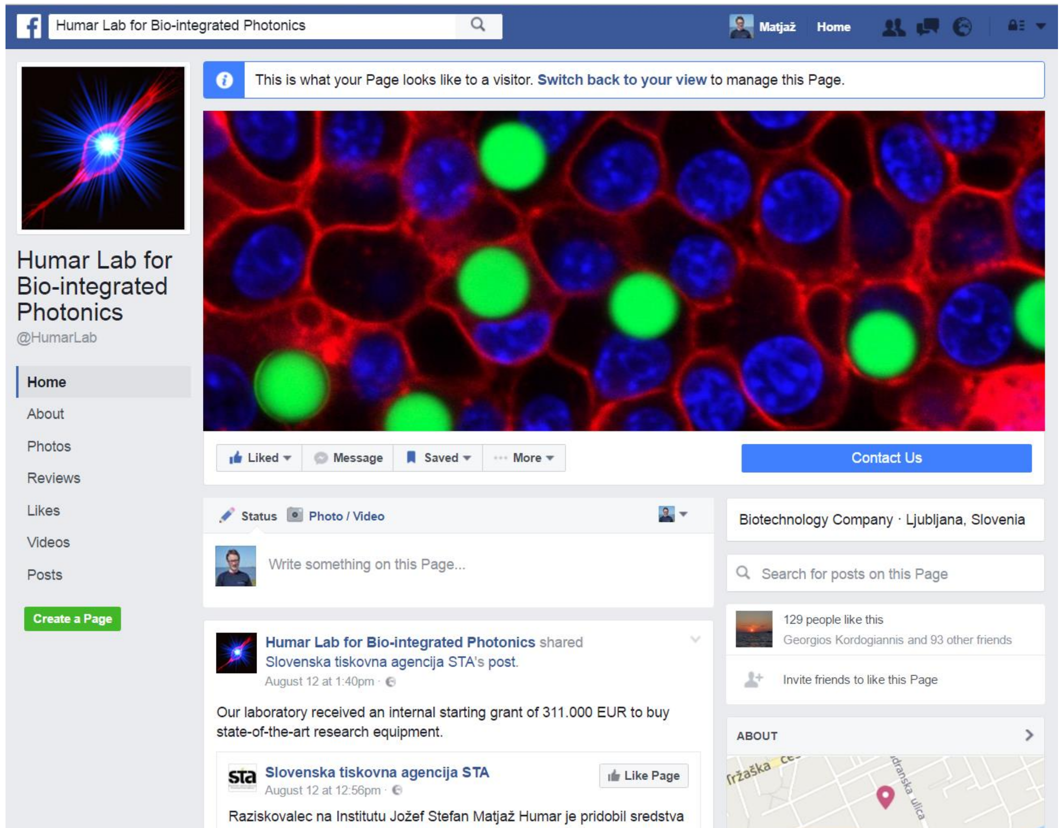

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