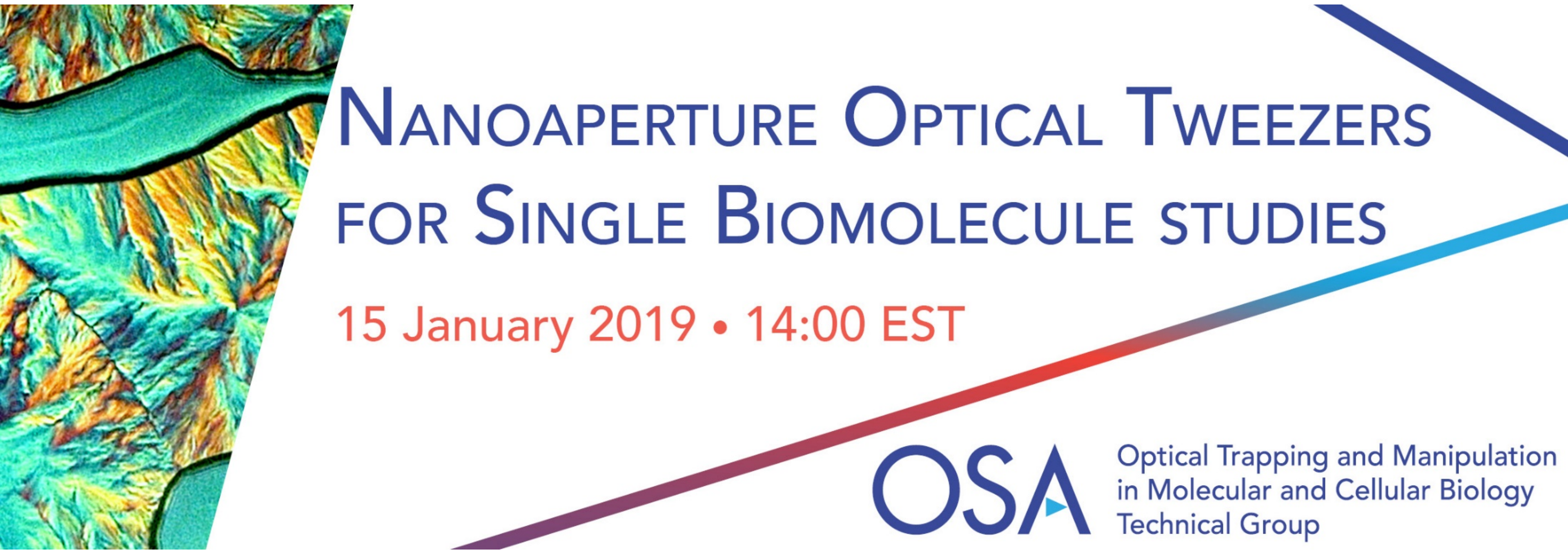
Nanoaperture Optical Tweezers for Single Biomolecule Studies

Presented by:

OSA
Optical Trapping and Manipulation in Molecular and Cellular Biology Technical Group

The OSA Optical Trapping and Manipulation in Molecular and Cellular Biology Technical Group Welcomes You!



OSA

Optical Trapping and Manipulation
in Molecular and Cellular Biology
Technical Group

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OSA
Optical Trapping and Manipulation
in Molecular and Cellular Biology
Technical Group

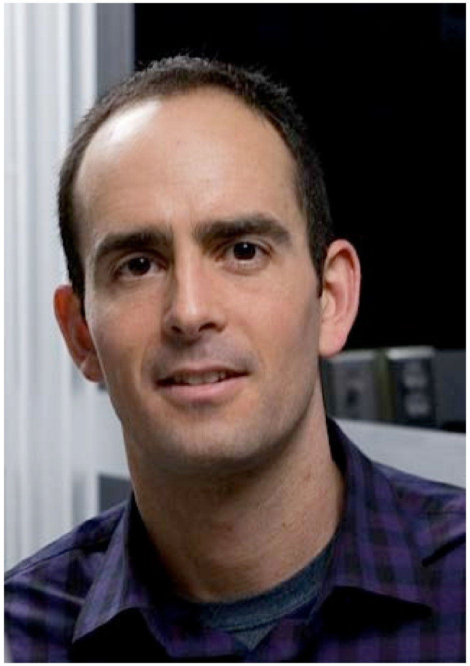
Technical Group at a Glance

- Focus
  - Development and application of novel optical trapping and manipulation techniques to biological problems
  - **630** members
- Mission
  - To benefit ***YOU***
  - Webinars, technical events, network events
  - Interested in presenting your research? Have ideas for technical group events? Contact us!
- Find us here
  - Website: www.osa.org/BT
  - Twitter: hashtag #BTTechGroupOSA

OSA Optical Trapping and Manipulation in Molecular and Cellular Biology Technical Group

Today's Webinar:

Nanoaperture Optical Tweezers for Single Biomolecule Studies

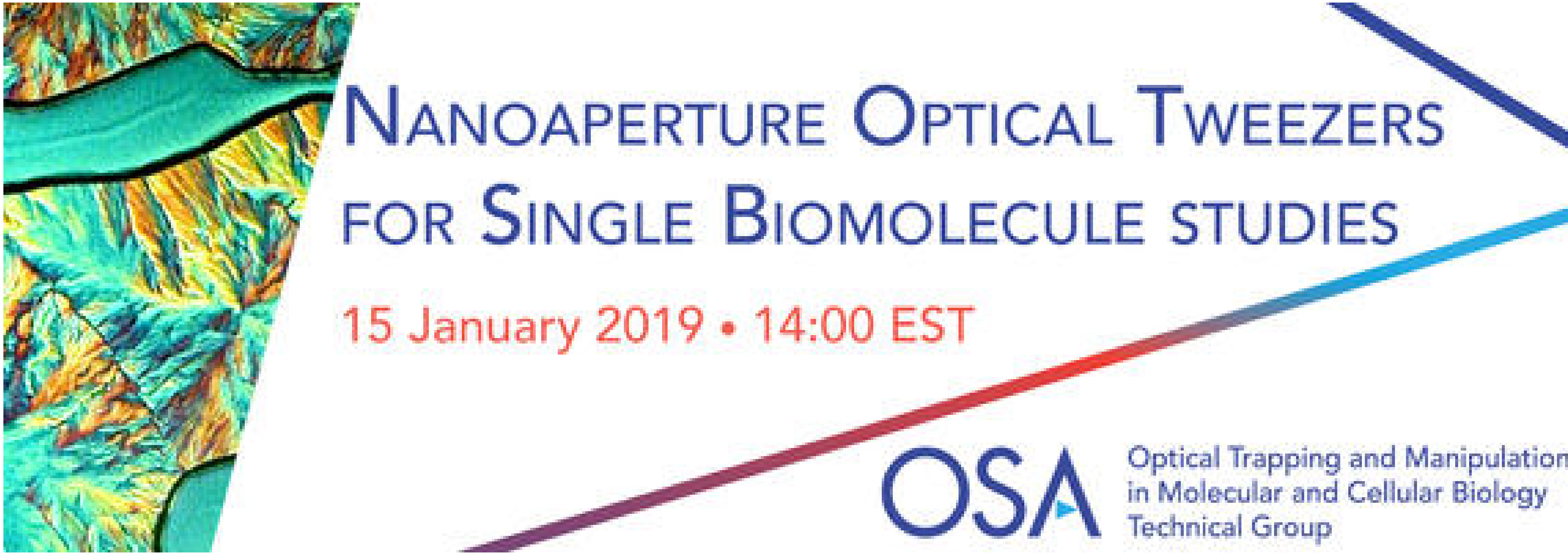


Prof. Reuven Gordon

Research Group leader of Nanoplasmonics Research Lab
University of Victoria, Canada

Speaker's Short Bio:
B.S. and M.S. from University of Toronto, Canada
Ph.D. in Physics from the University of Cambridge, UK
Fellow of OSA (2016), SPIE (2018), and IEEE (2019)

OSA Optical Trapping and Manipulation in Molecular and Cellular Biology Technical Group



Reuven Gordon
University of Victoria, Canada

2018 Nobel Prize in Physics

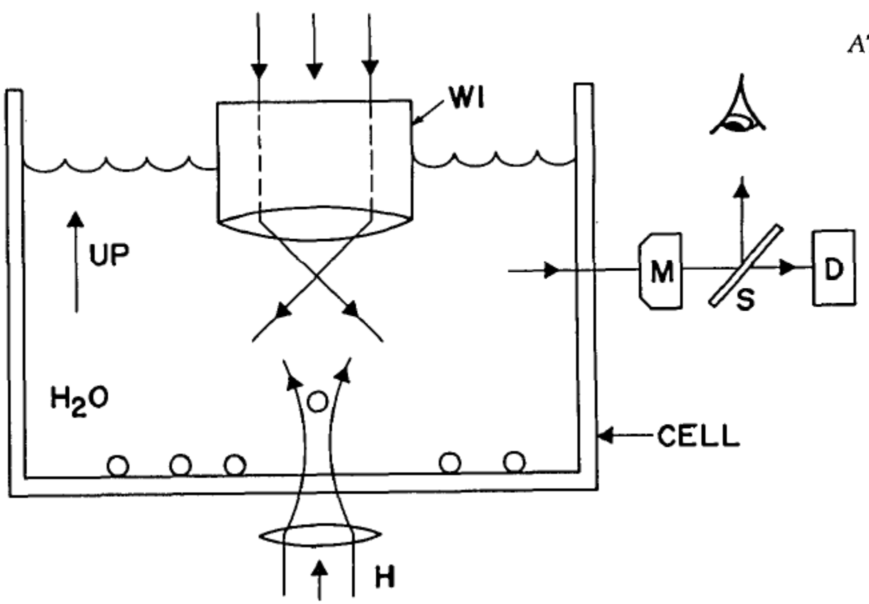
50% to Arthur Ashkin "for the optical tweezers and their application to biological systems"

288 OPTICS LETTERS / Vol. 11, No. 5 / May 1986

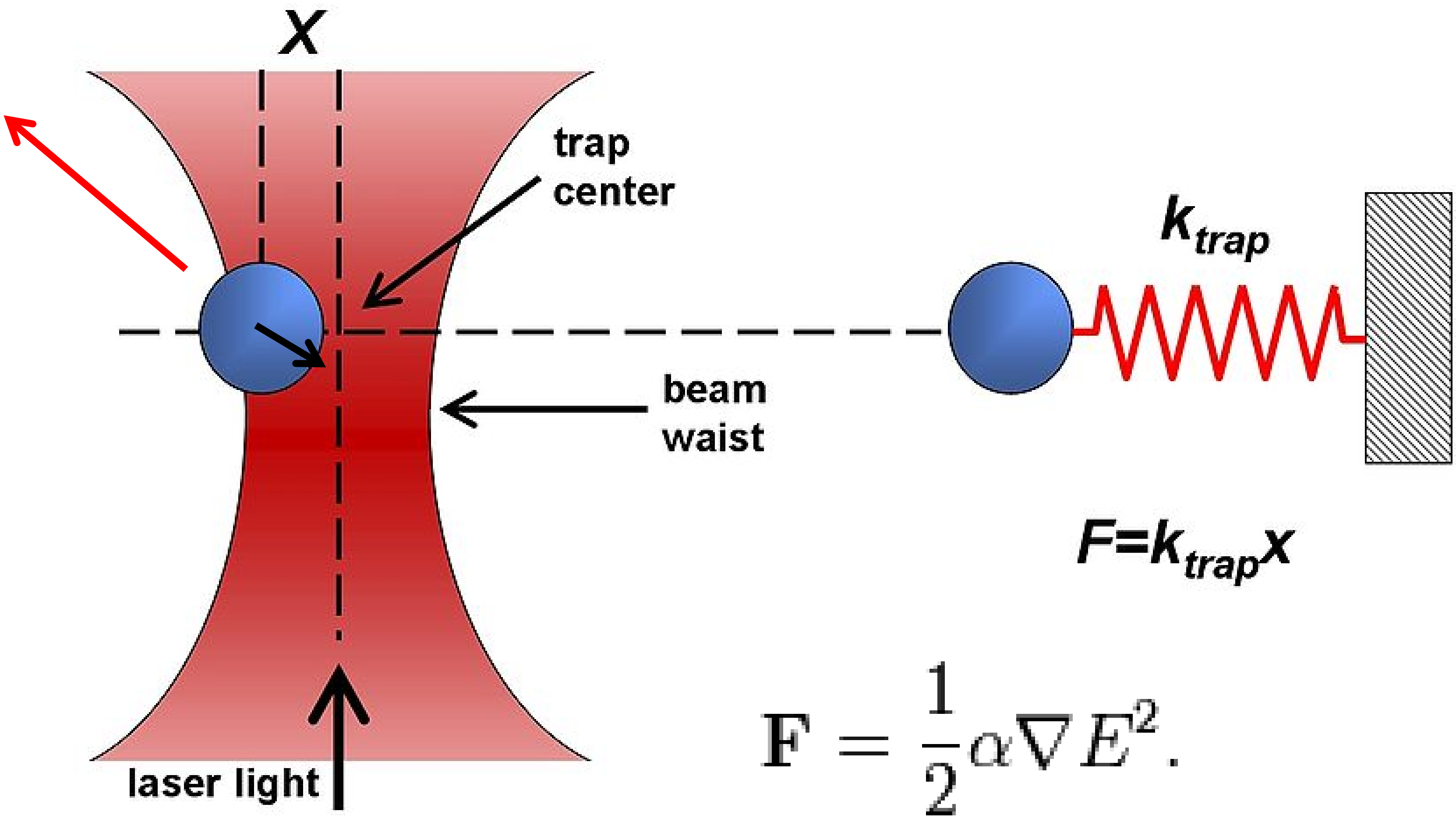
Observation of a single-beam gradient force optical trap for dielectric particles

A. Ashkin, J. M. Dziedzic, J. E. Bjorkholm, and Steven Chu

AT&T Bell Laboratories, Holmdel, New Jersey 07733



Optical Trapping

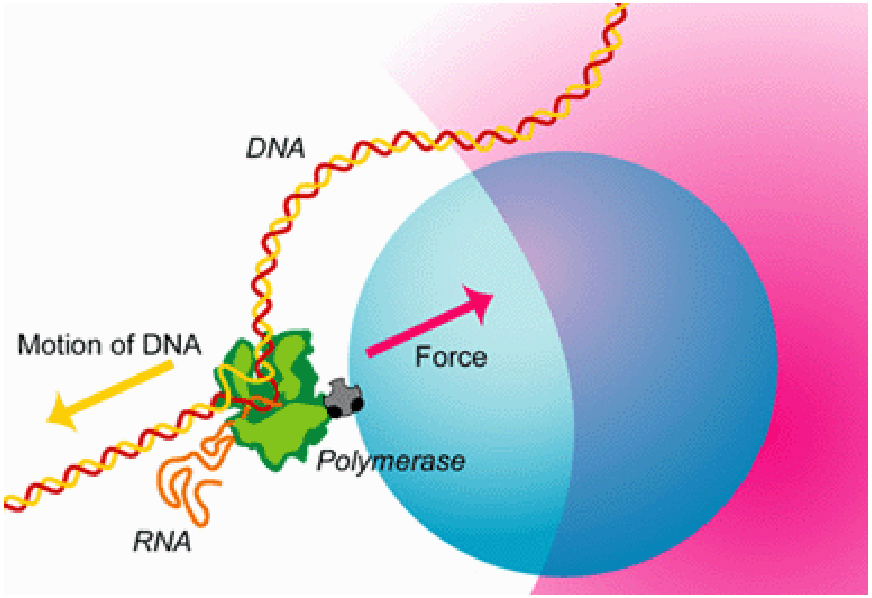


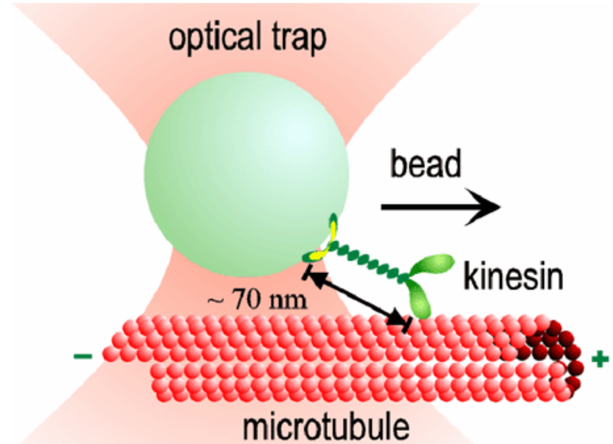
$$\mathbf{F} = \frac{1}{2}\alpha\nabla E^2.$$

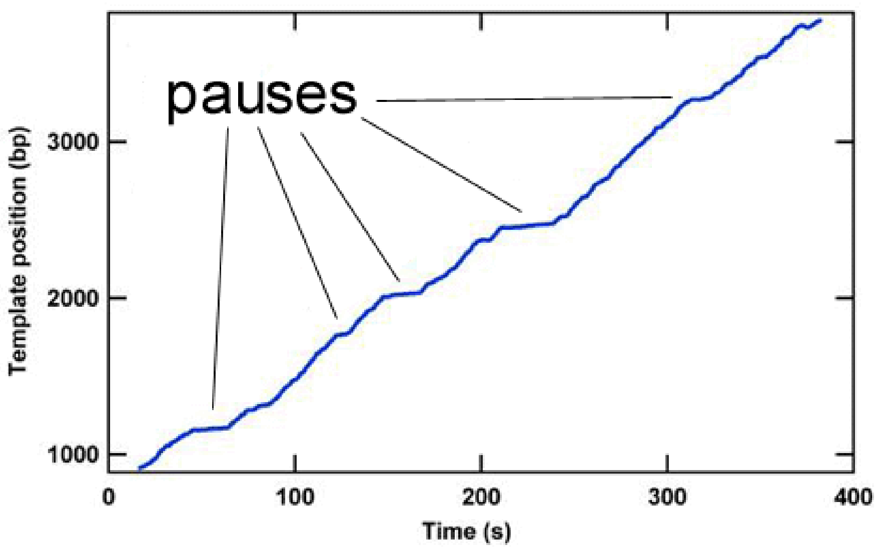
$$\alpha \sim a^3$$

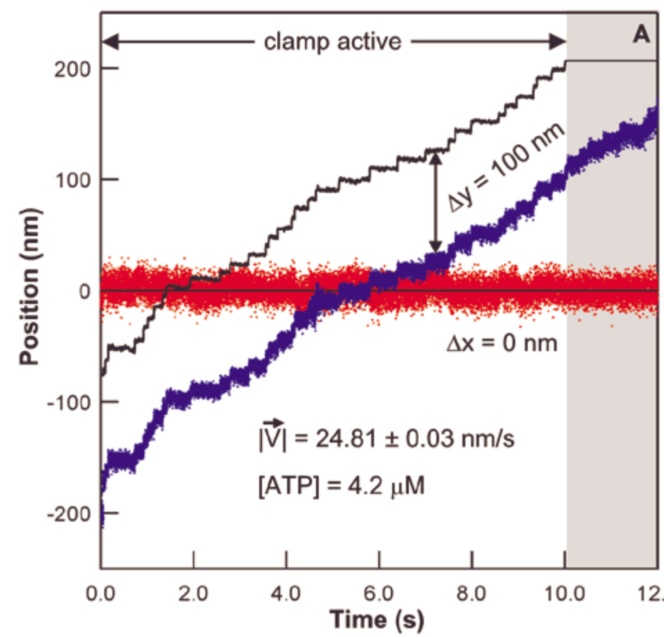
https://en.wikipedia.org/wiki/Optical_tweezers

Some Bio Examples (Block Lab)



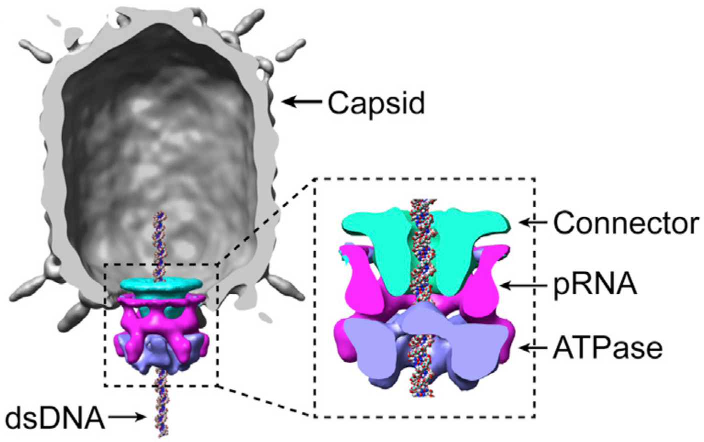


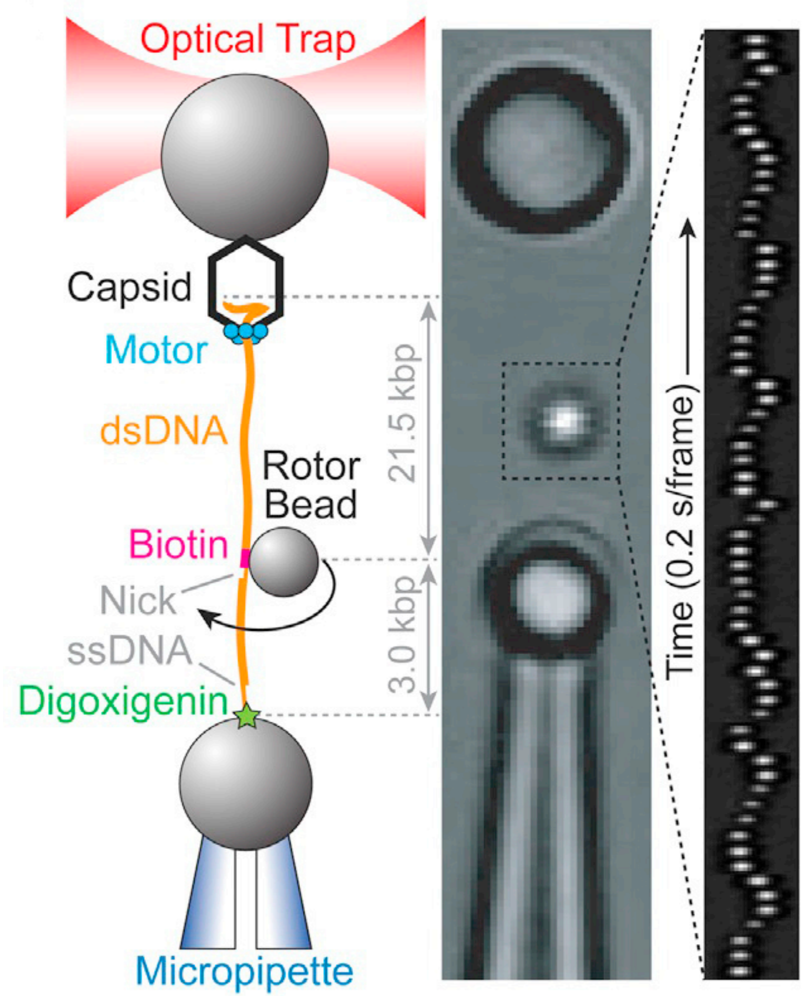




https://blocklab.stanford.edu/research.html

More Examples (Bustamante Lab)



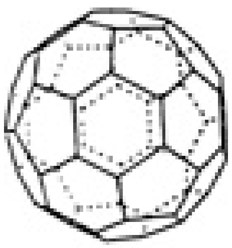


https://phys.org/news/2014-05-viral-packaging-motor-rotates-dna.html

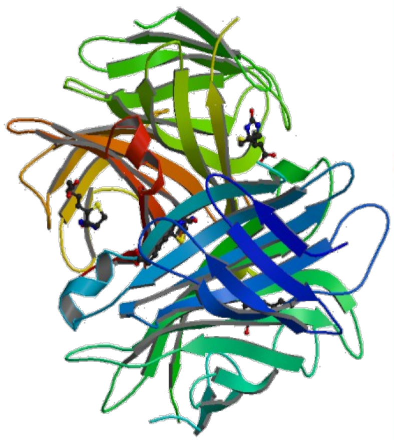
Reflections

- Proteins are the machines of life
- Tweezers allow us to observe their action, but require large tethers
- Can we hold onto single proteins?

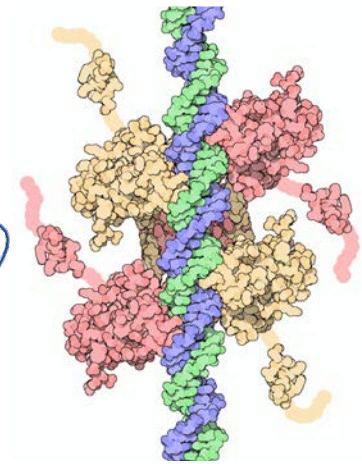
Nano-Bio: 1 nm to 50 nm



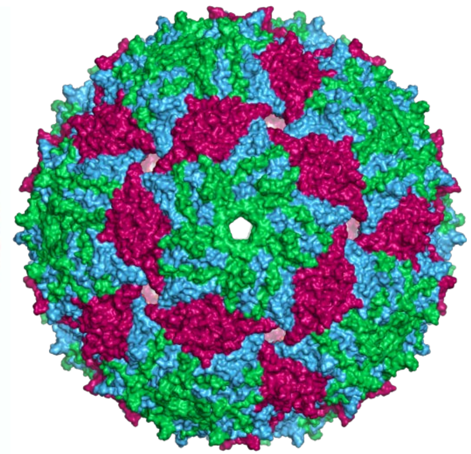
C60
0.7 nm
(too small?)



Proteins,
Complexes,
Interactions
~1-5 nm



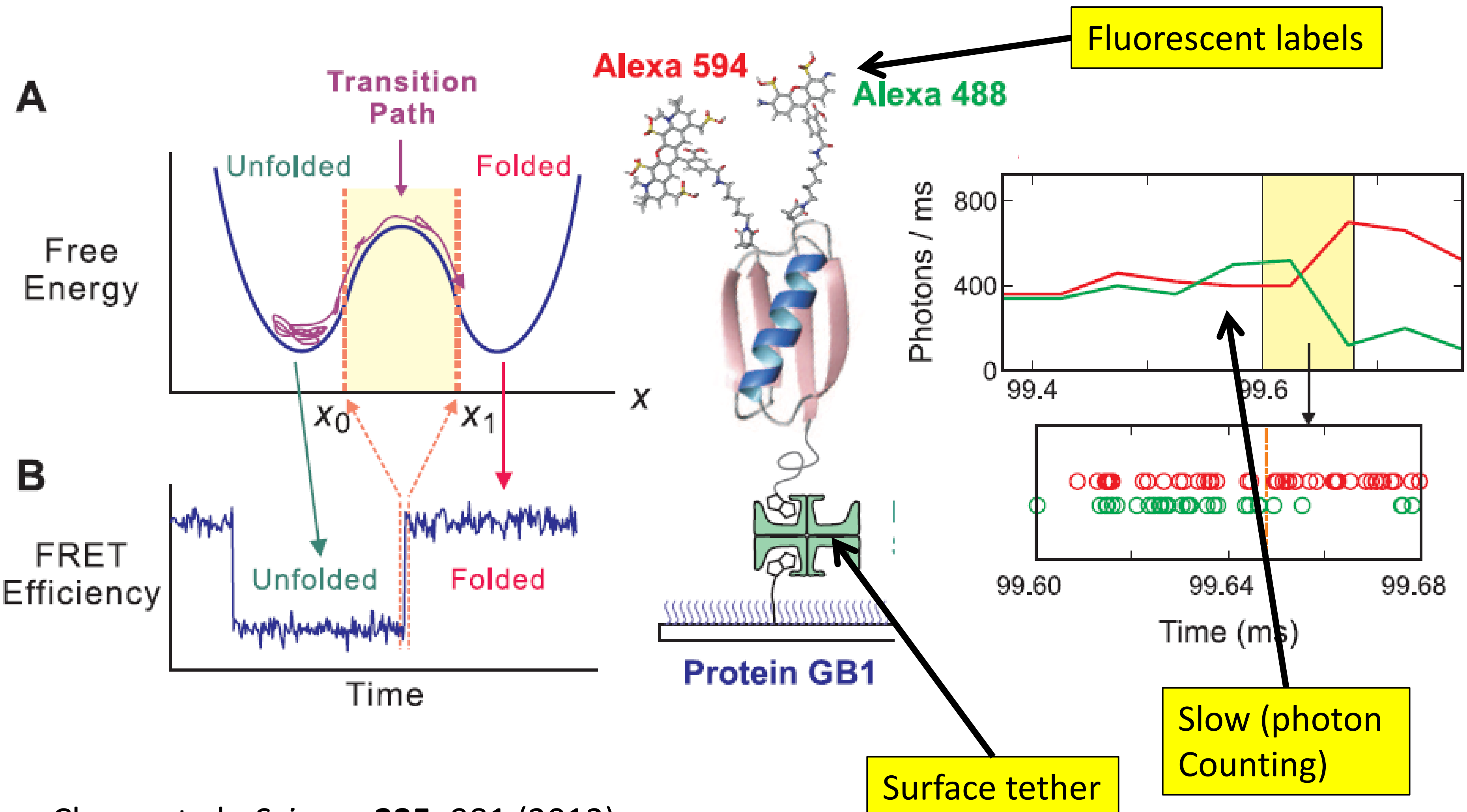
DNA
Protein-DNA
~1-10 nm



Virion
MS2
27 nm

…

Single Molecule Protein Folding Study



Chung et al., *Science* **335**, 981 (2012)

Single molecule studies

Fluorescence (etc.)

- Fluorescent labels
  - Slow (~ms)
  - Blinking
  - Bleaching (limited observation)
  - Tags alter structure
- Tether
  - Alters structure
  - Hinders motion, blocks sites
- Indirect measurement
  - FRET open to interpretation

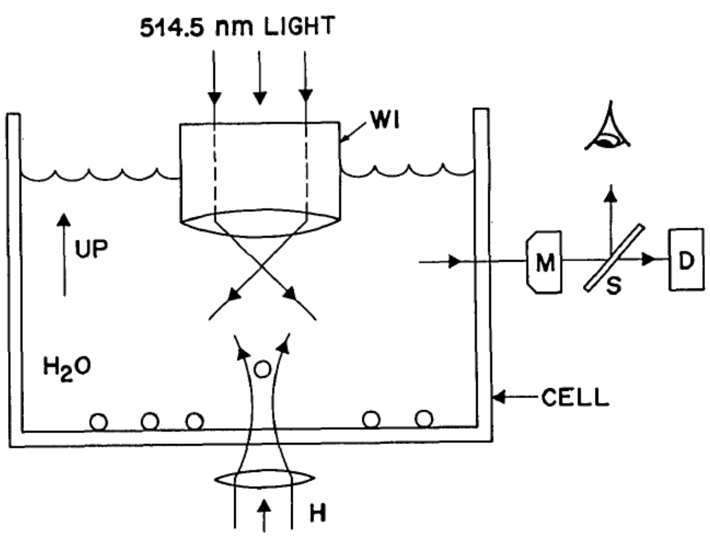
Double Nanohole Optical Tweezer

- Label free
  - Fast (~ns)
  - Intrinsic
  - Extended observation period
- Free solution
  - No tethering
  - Free to move around in trap
- Direct measurement
  - Elastic light scatter is a measure of the molecule's polarizability

Observation of a single-beam gradient force optical trap for dielectric particles

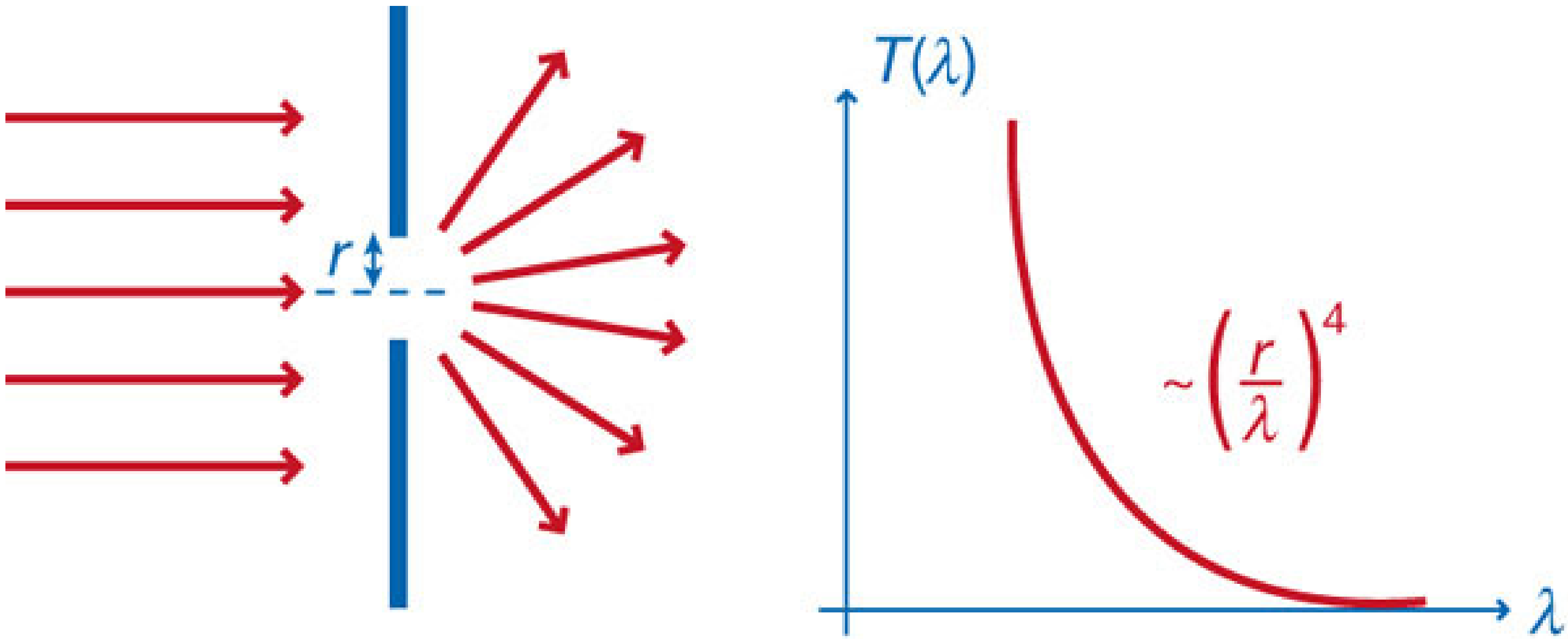
A. Ashkin, J. M. Dziedzic, J. E. Bjorkholm, and Steven Chu

AT&T Bell Laboratories, Holmdel, New Jersey 07733



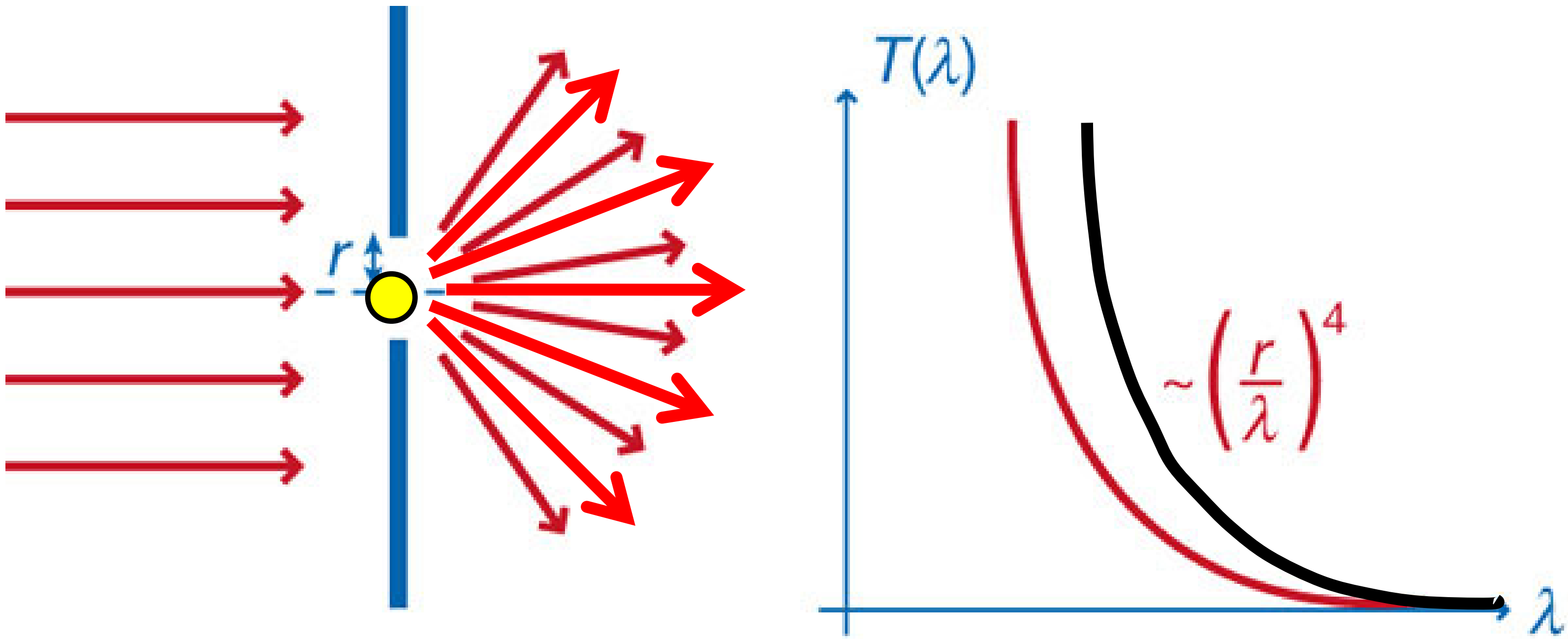
Additional experiments were performed on individual colloidal polystyrene latex particles in water. Unfortunately the particles exhibit a form of optical damage at high optical intensities. For 1.0-μm spheres with a trapping power of a fraction of a milliwatt, particles survived for tens of minutes and then shrank in size and disappeared. Spheres of 0.173 μm were trapped for several minutes with a power of a milliwatt before being lost. Particles of 0.109-μm diameter required about 12–15 mW and survived about 25 sec. With 85- and 38-nm latex particles the damage was so rapid that it was difficult to observe the scattering reliably. It was nevertheless clear that trapping occurred over full size range from Mie to Rayleigh particles.

Optical Trapping with Nanoholes



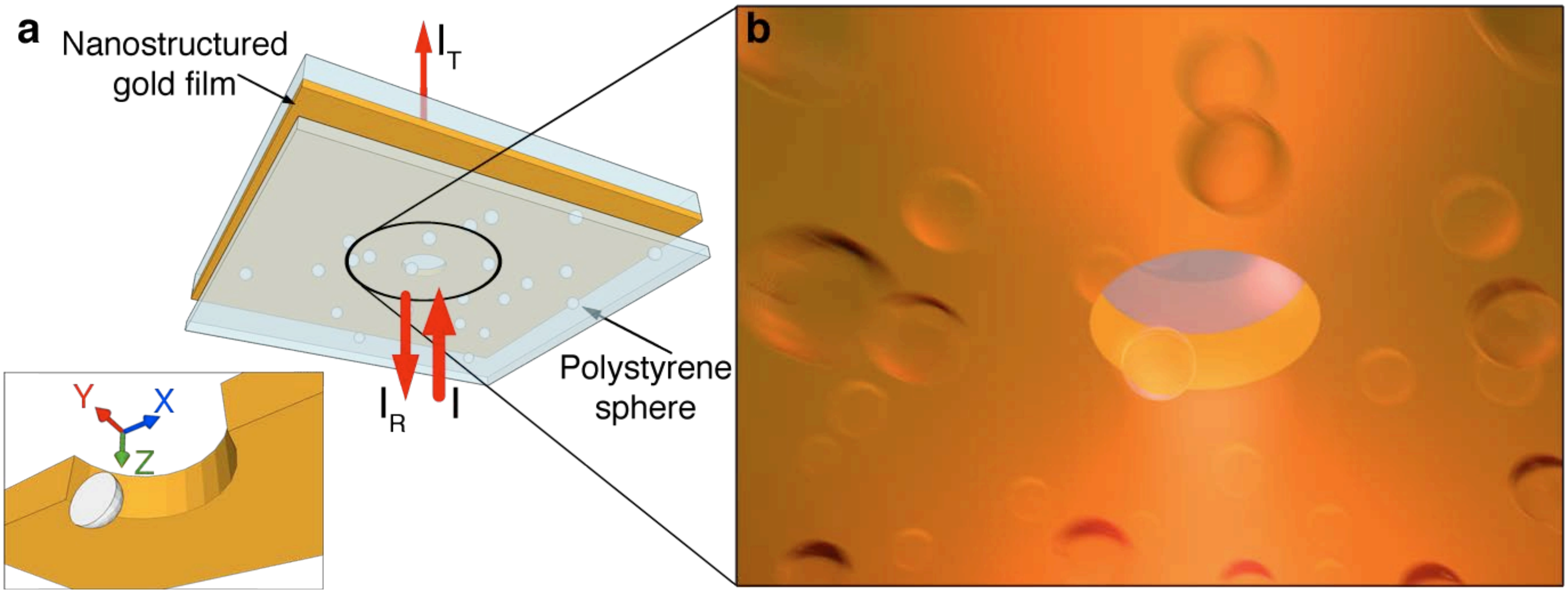
Play off a third power scaling law against fourth power scaling law.

Optical Trapping with Nanoholes



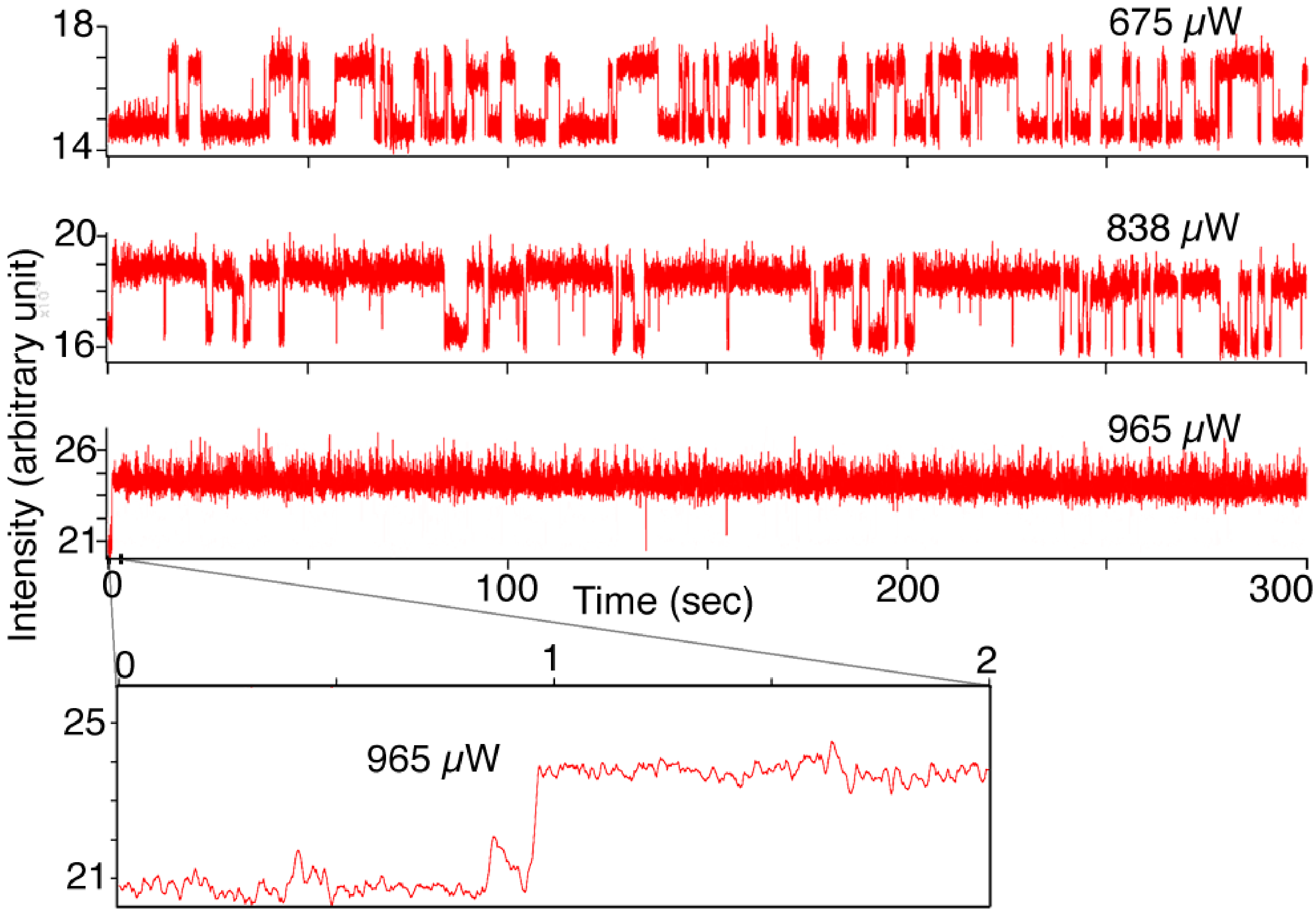
Play off a third power scaling law against fourth power scaling law.

Experiment

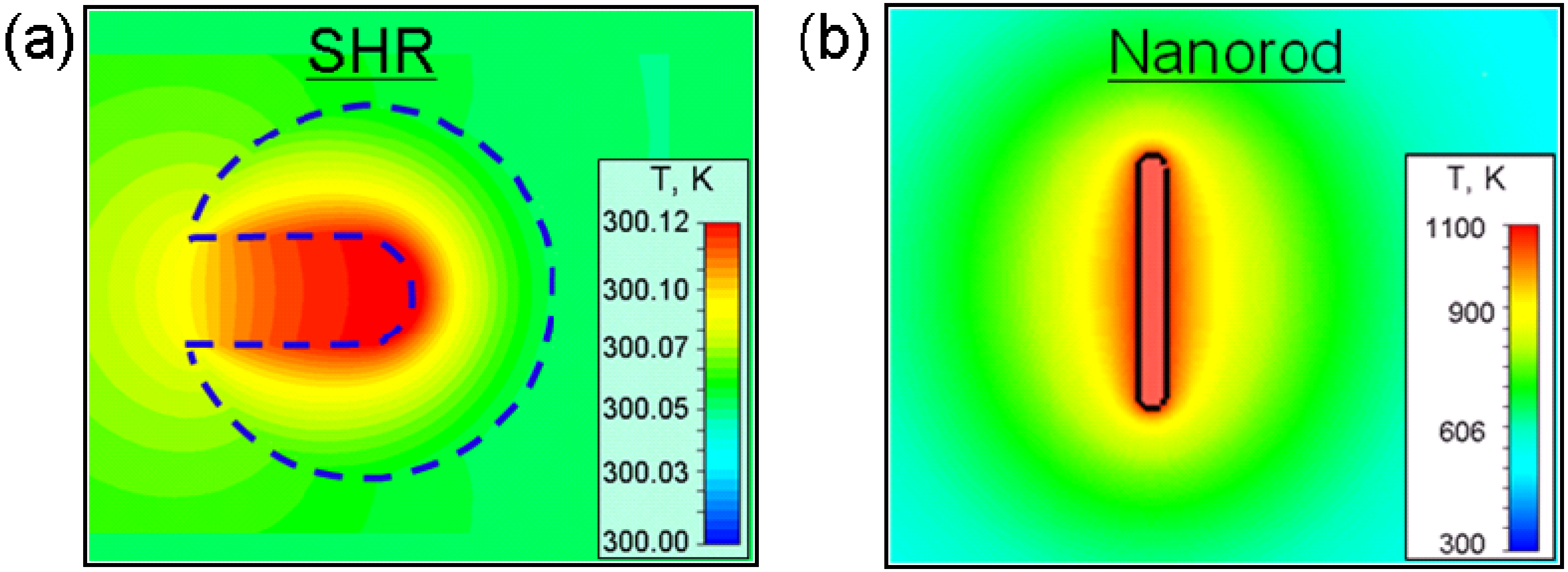


M. L. Juan, R.Gordon, Y. Pang, F. Eftekhari, R. Quidant, "Self-induced back-action optical trapping of dielectric nanoparticles," *Nature Physics,* 5, 915-919 (2009).

Trapping Events @ 100 nm



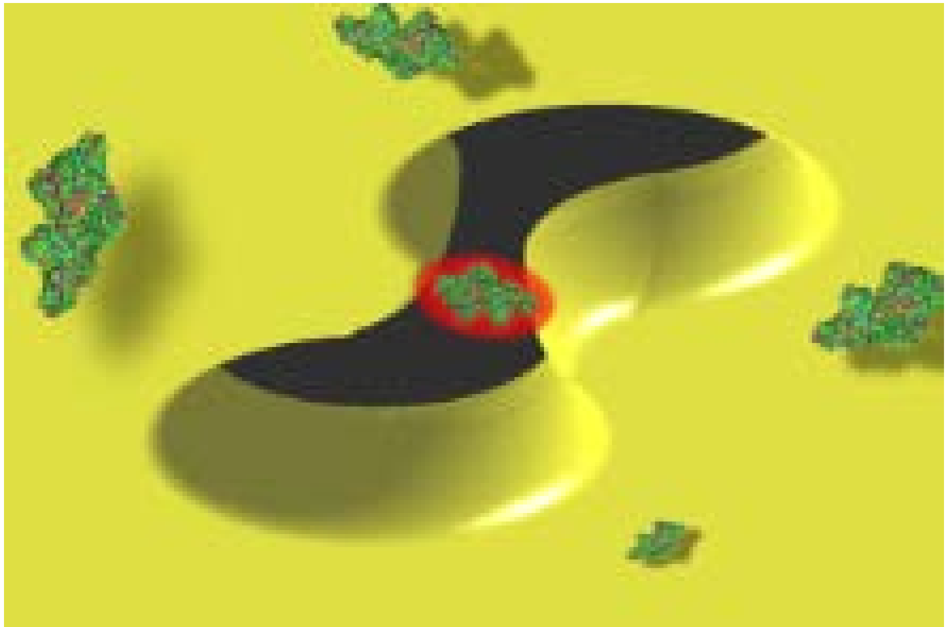
Low heating



Pavel N. Melentiev, Anton E. Afanasiev, Artur A. Kuzin, Andrey S. Baturin, and Victor I. Balykin, "Giant optical nonlinearity of a single plasmonic nanostructure," Opt. Express 21, 13896-13905 (2013)

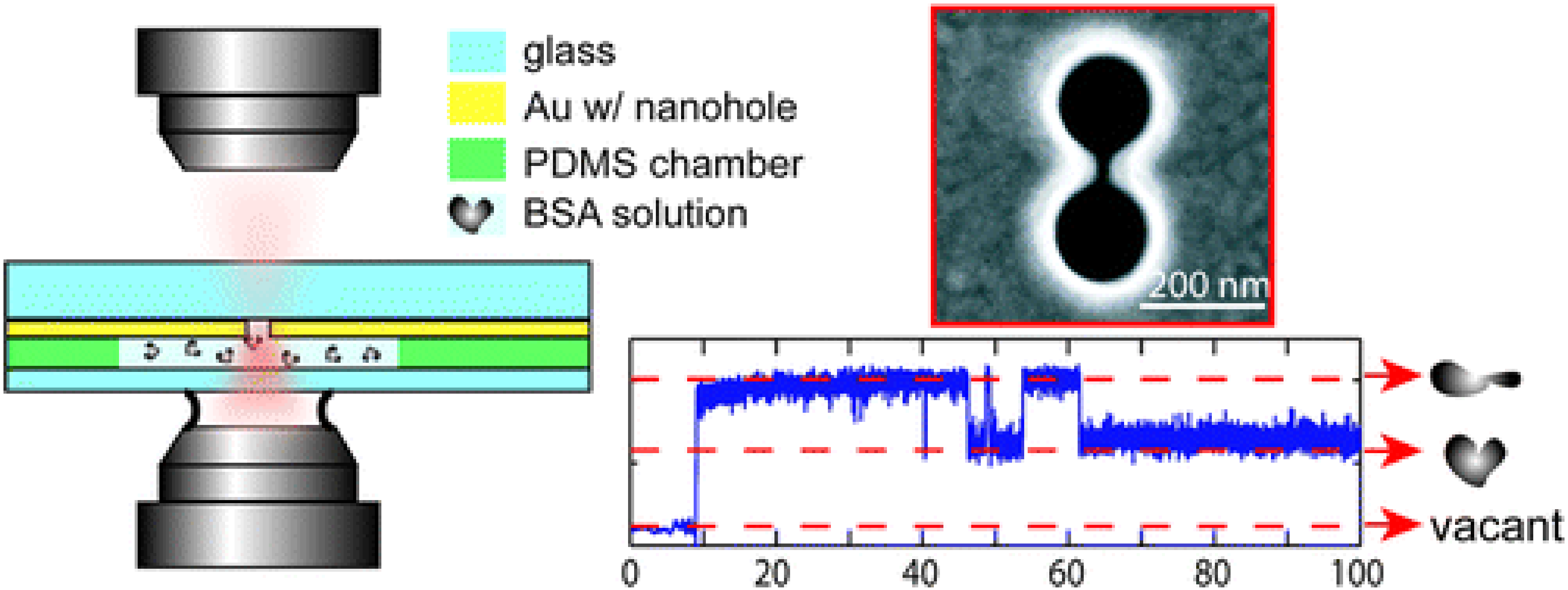
Earlier Works

- Theoretical work on aperture trapping calculated micron bead trapping with 100 nm apertures [*Phys. Rev. Lett.* **1999,** 83, 4534]
- Trapping of 200 nm PS particles with high field gradients around apertures [*J. Phys. Chem. B* **2004,** *108,* 13607-13612]



DOUBLE NANOHOLE OPTICAL TWEEZERS

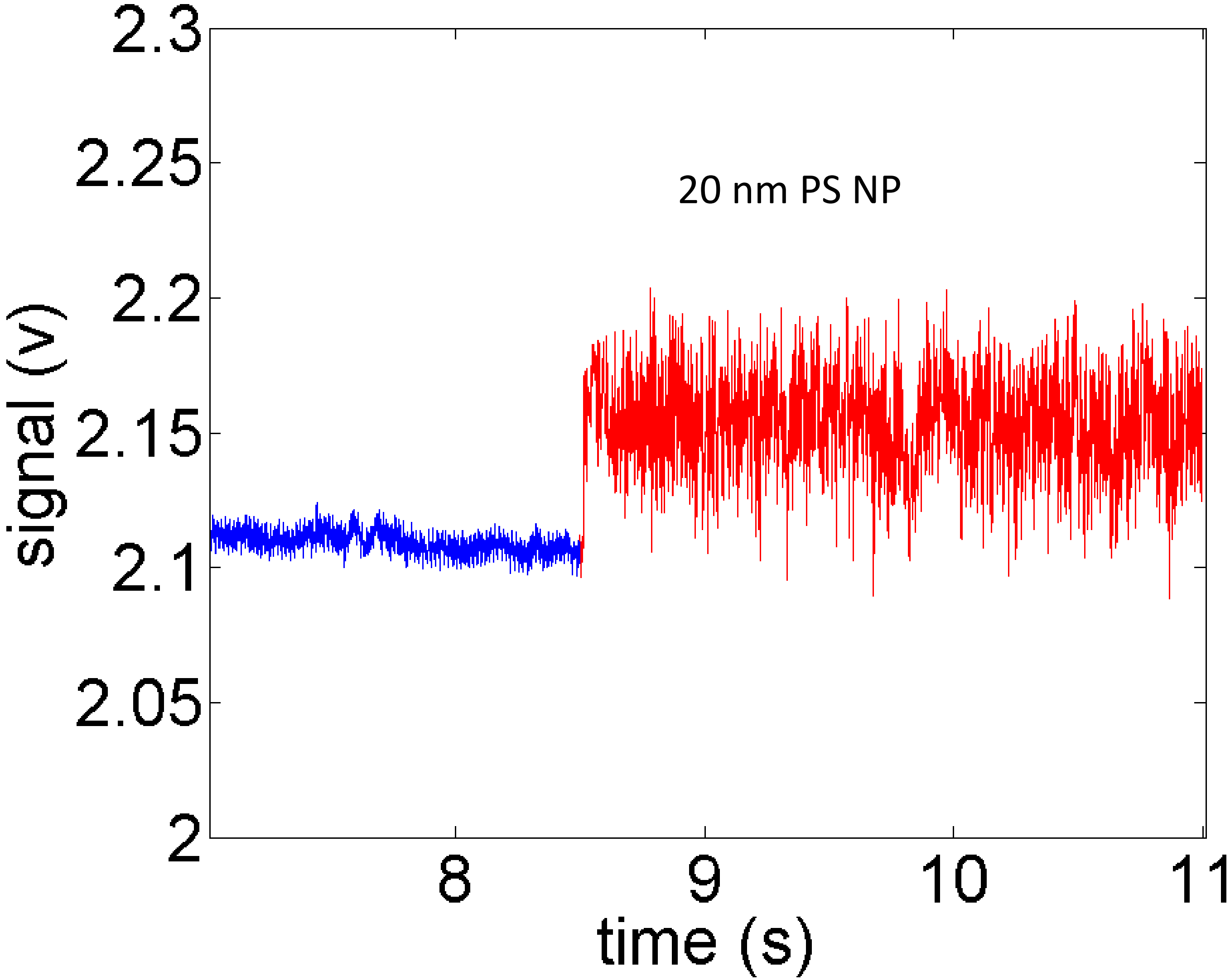
Single Protein Optical Trapping (+Sensing +Manipulation)



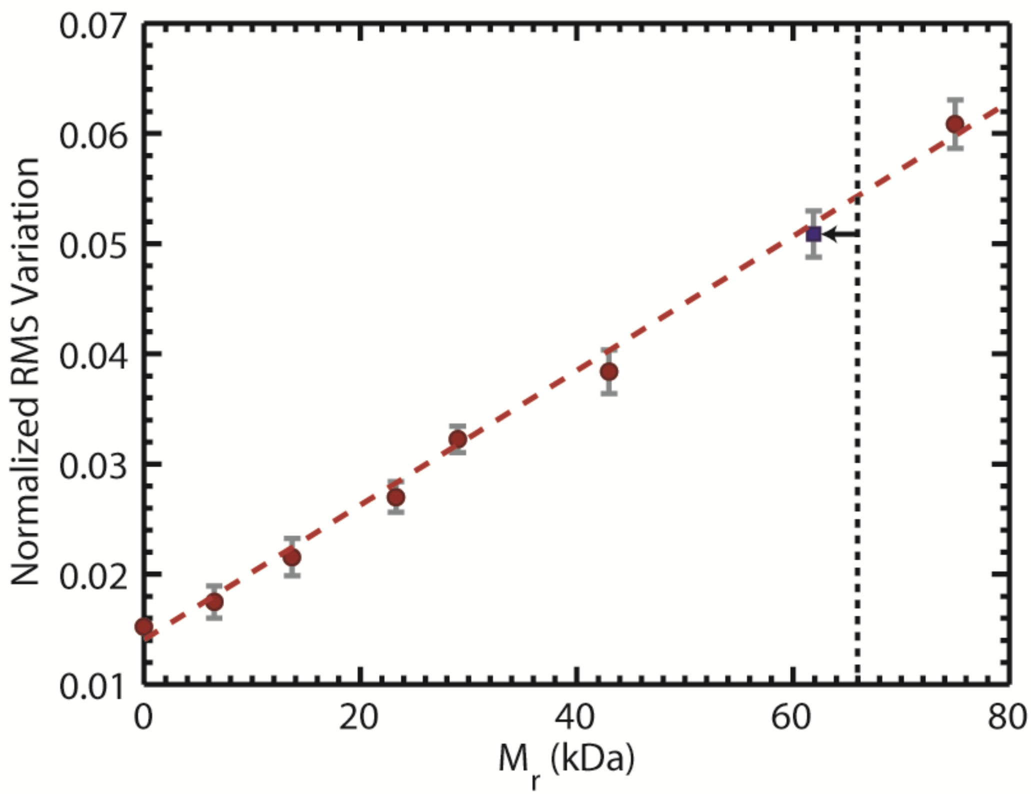
Demonstrated trapping and unfolding of a **single** protein – hydrodynamic radius of 3.4 nm. SNR = 33 at 1 kHz – **ultrasensitive**.

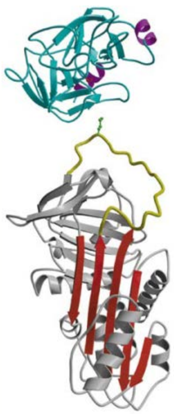
Y. Pang, R. Gordon, "Optical trapping of a single protein," *Nano Letters* , 12(1), 402-406 (2012).

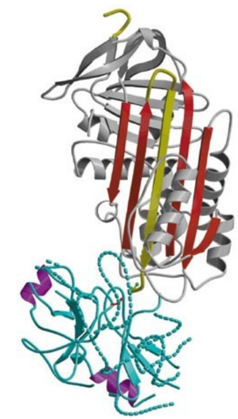
“Noise” in Trapping



Protein Sizing from Standard Deviation of Noise

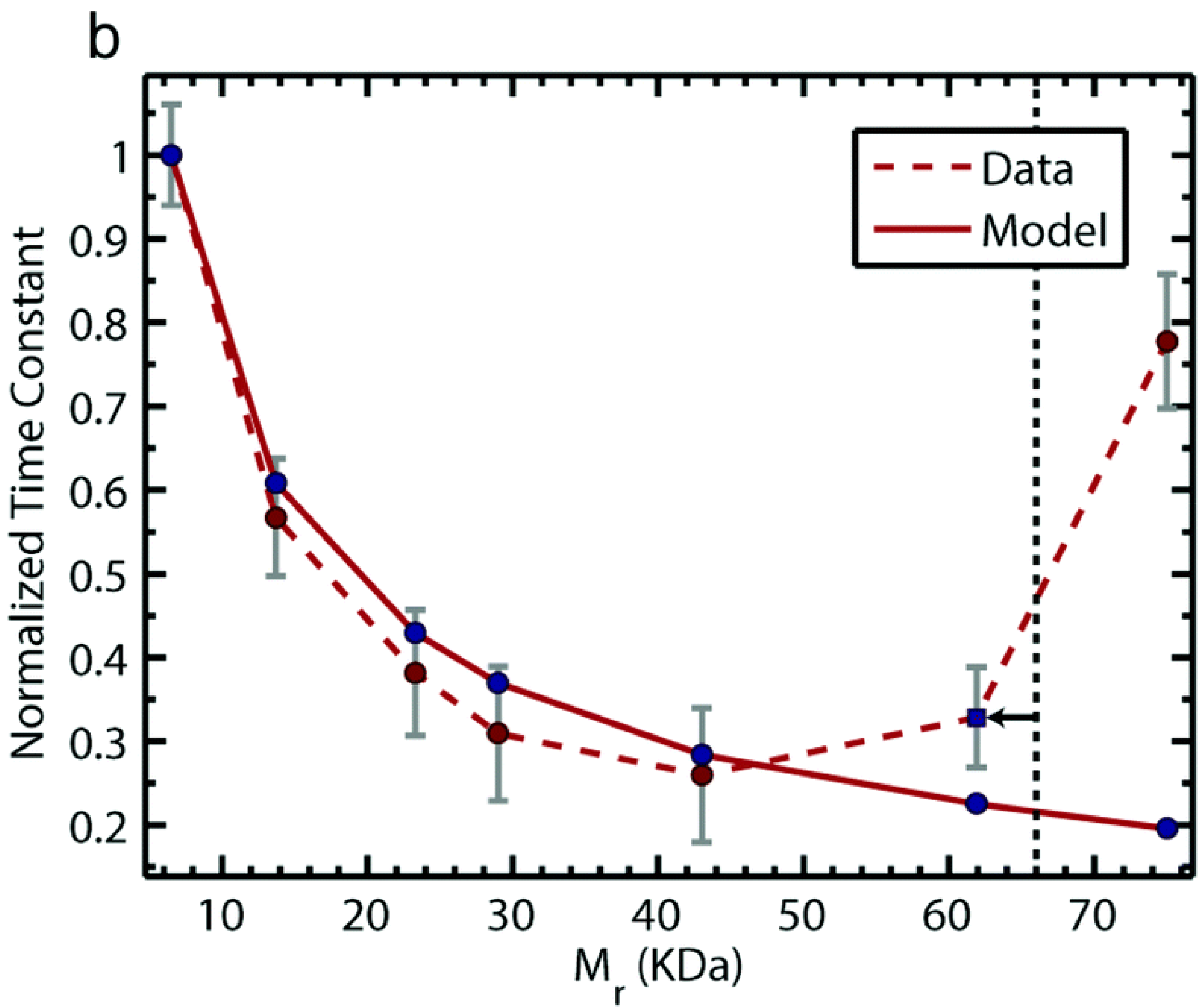




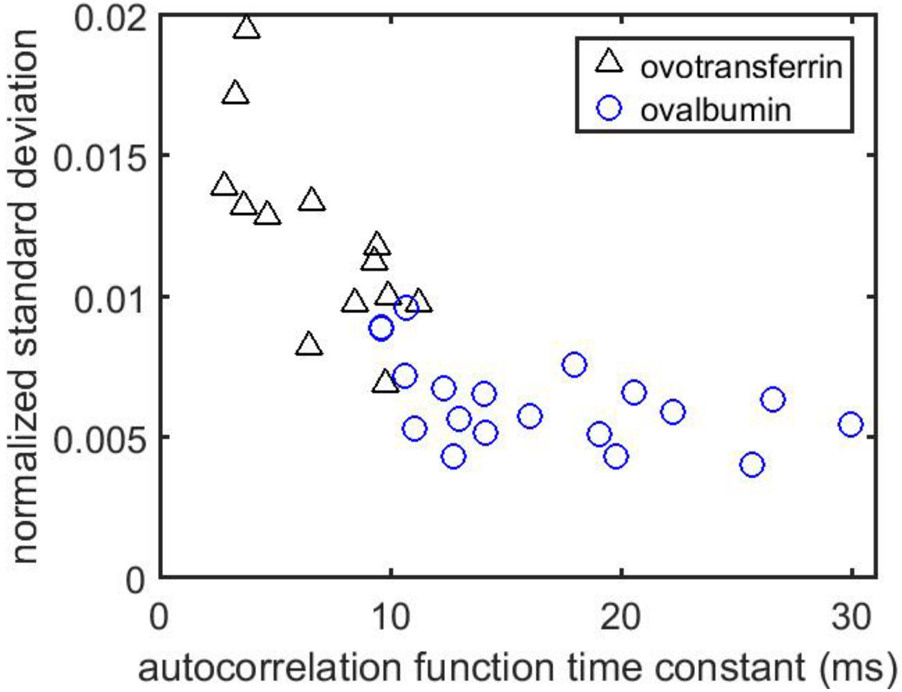


S. Wheaton, R. Gordon, “Molecular weight characterization of single globular proteins using optical nanotweezers”, Analyst, 140, 4799 - 4803 (2015).

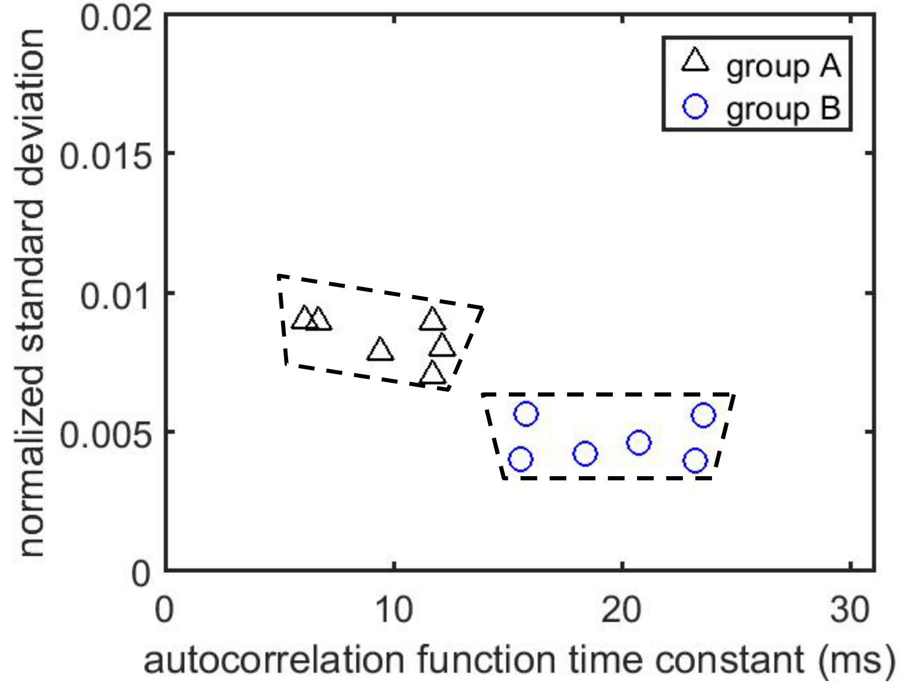
Autocorrelation Time Constant



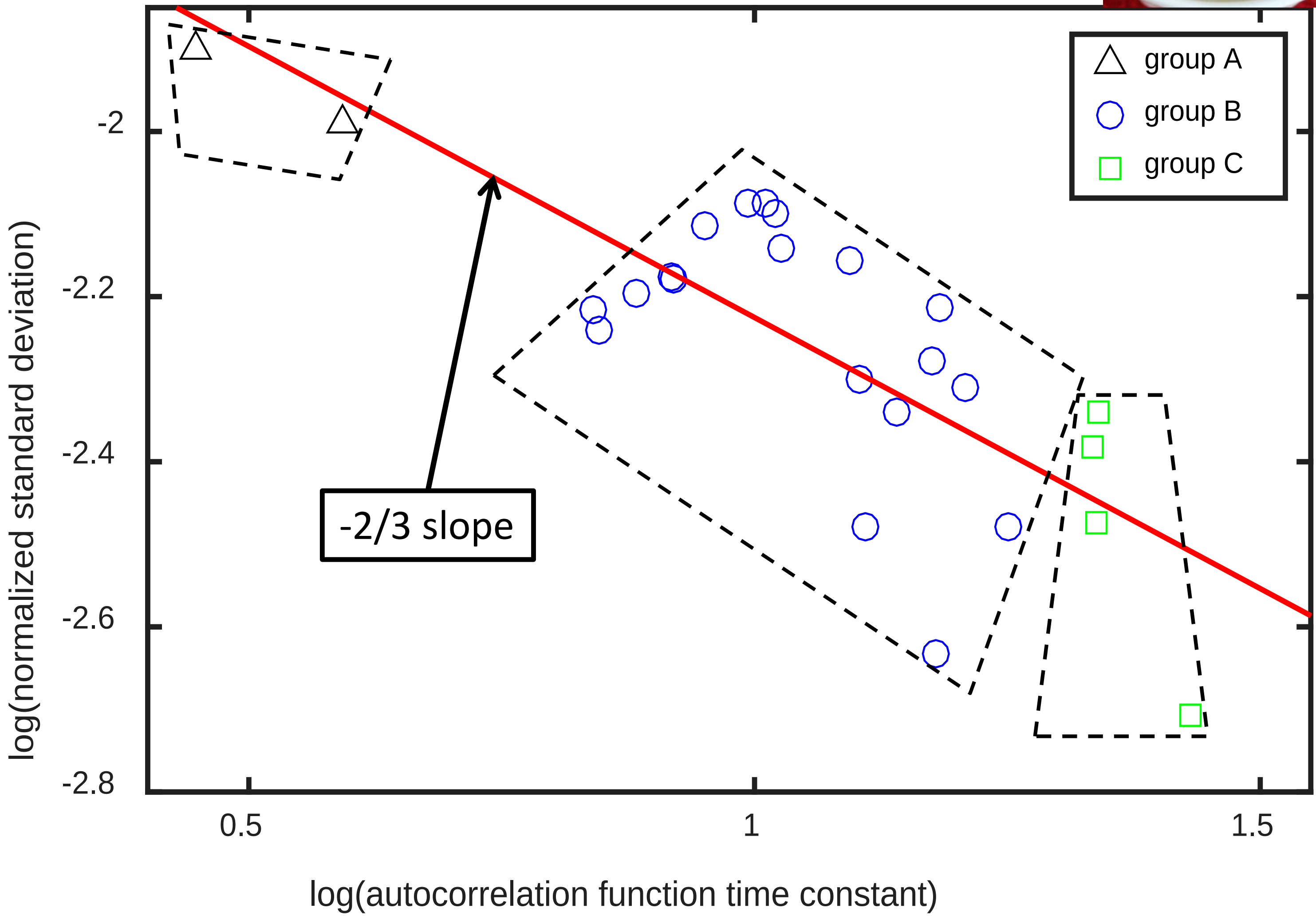
Studying Heterogeneous Samples



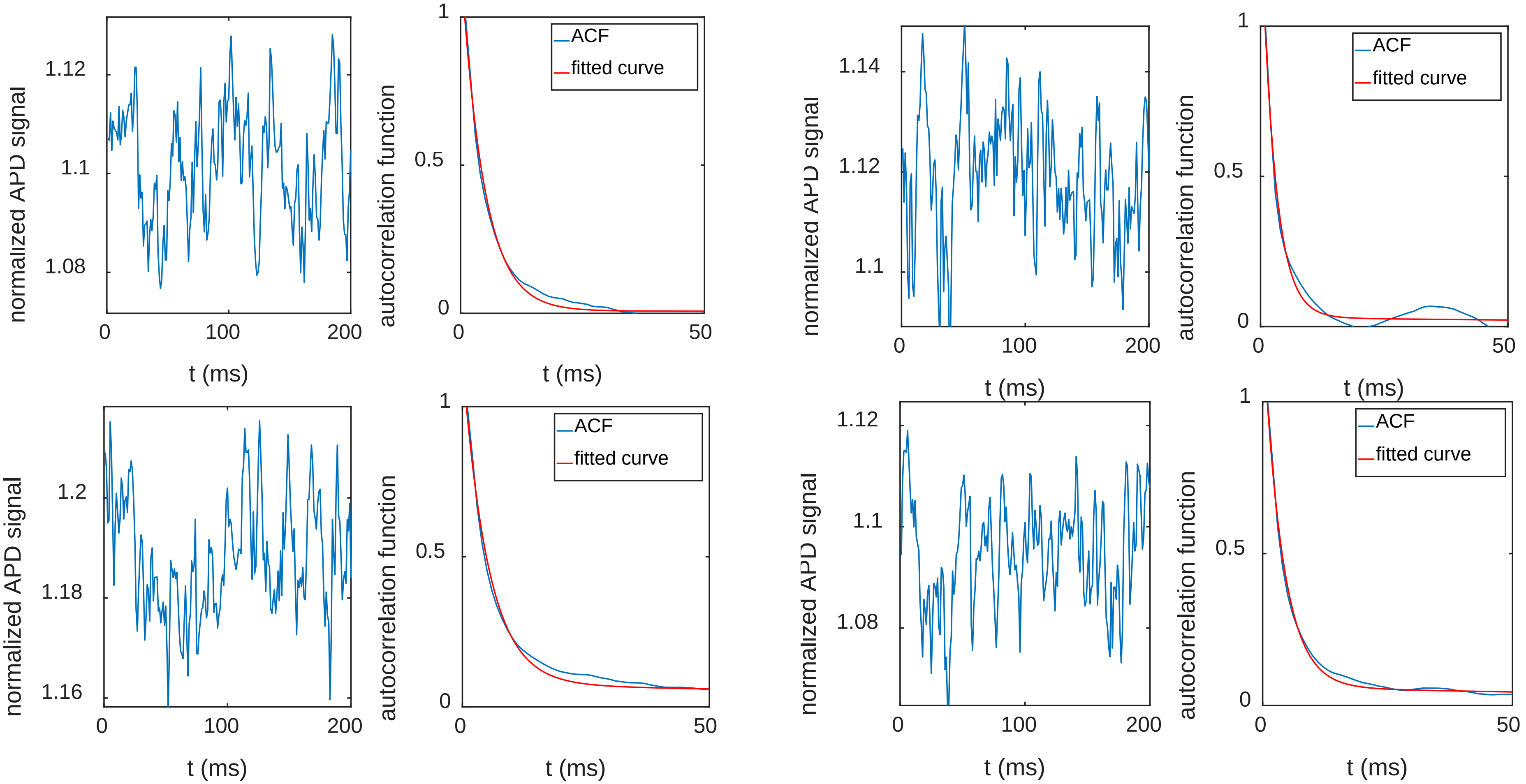
Equal mixture



Egg White Sample

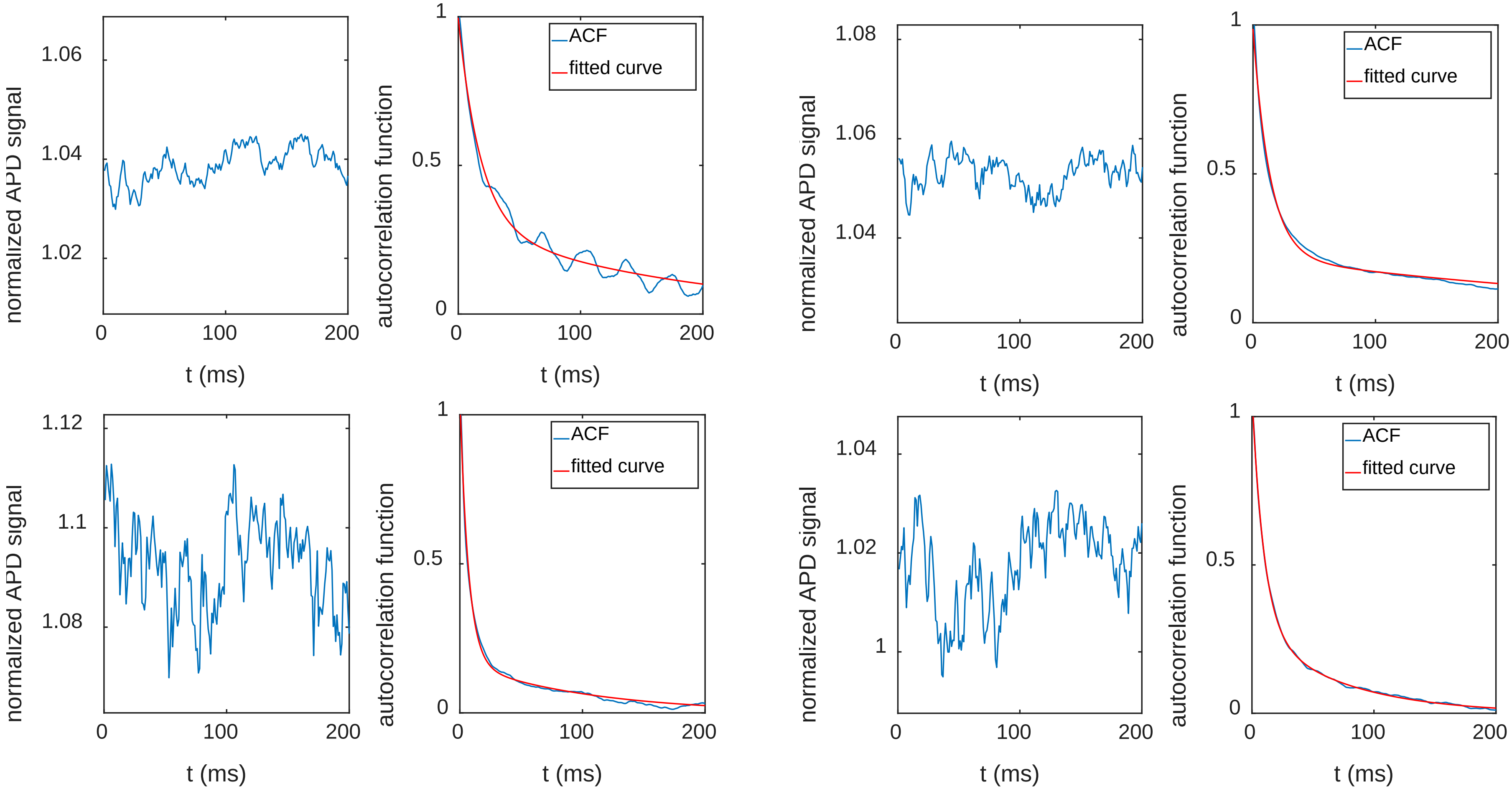


Ovotransferrin - Group A



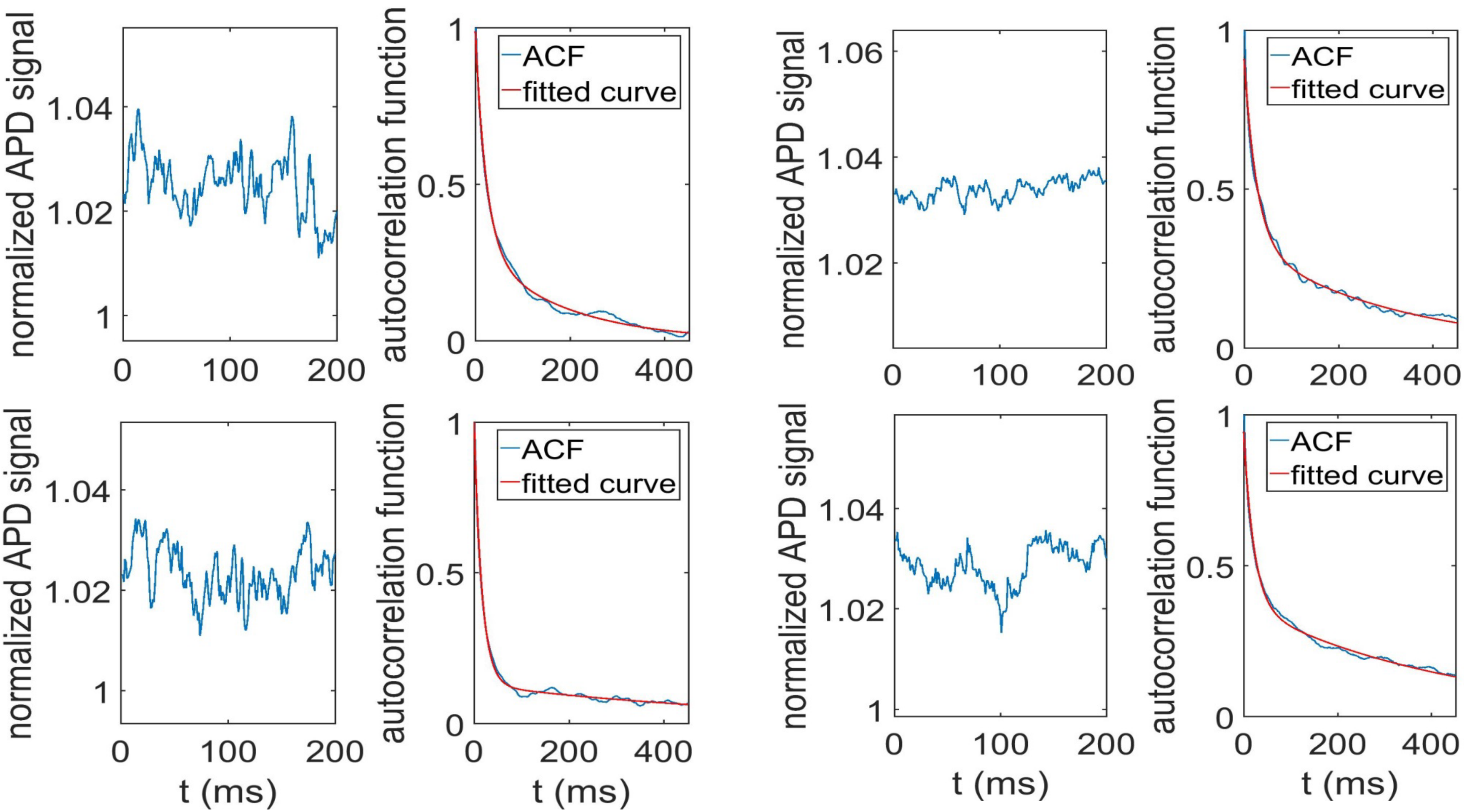
Fast time constant around 4 ms.

Ovalbumin – Group B



Fast time constant around 12 ms.

Ovomucoid – Group C



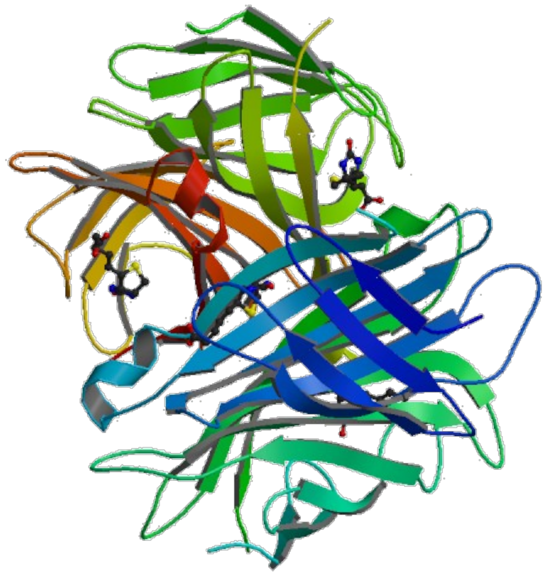
Fast time constant around 22 ms.

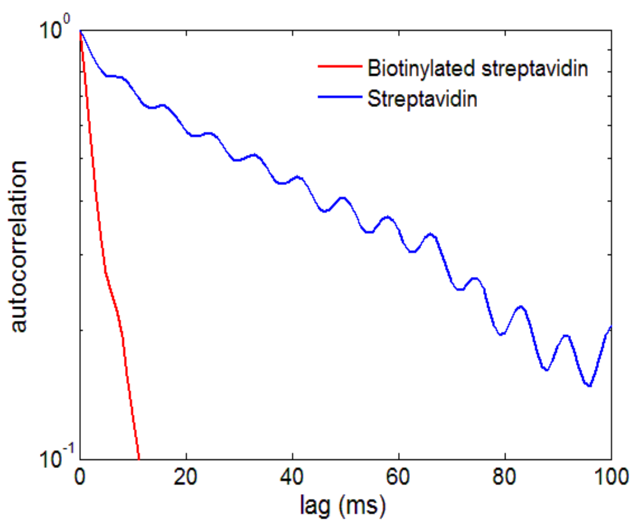
Composition Summary

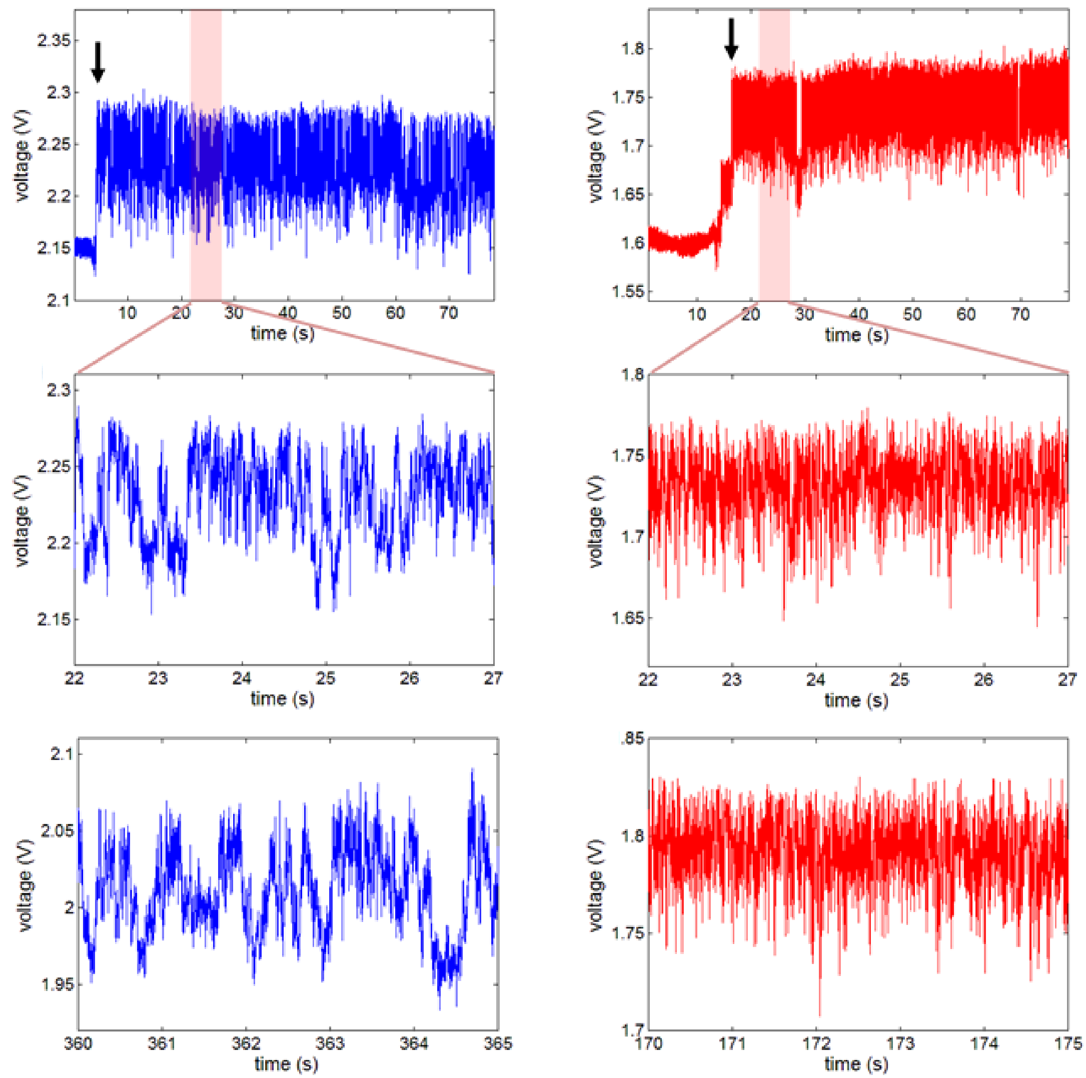
Experiment				Reference			
Group	M_r range (kDa)	Number of events	%	Protein name	M_r (kDa)	%	
A	$49 < M_r$	2	8%	ovotransferrin	78	13.5%	13.5%
				ovoglobulin G3	49	4.5%	
B	$36 < M_r < 49$	19	76%	ovalbumin	45	61%	70%
				ovoglobulin G2	36	4.5%	
C	$M_r < 36$	4	16%	ovomucoid	28	12.5%	16.5%
				lysozyme	14.3	4%	

N. Hacohen, C. J. X. Ip, R. Gordon, "Analysis of Egg White Protein Composition with Double Nanohole Optical Tweezers," ACS Omega 3, 5266-5272 (2018).

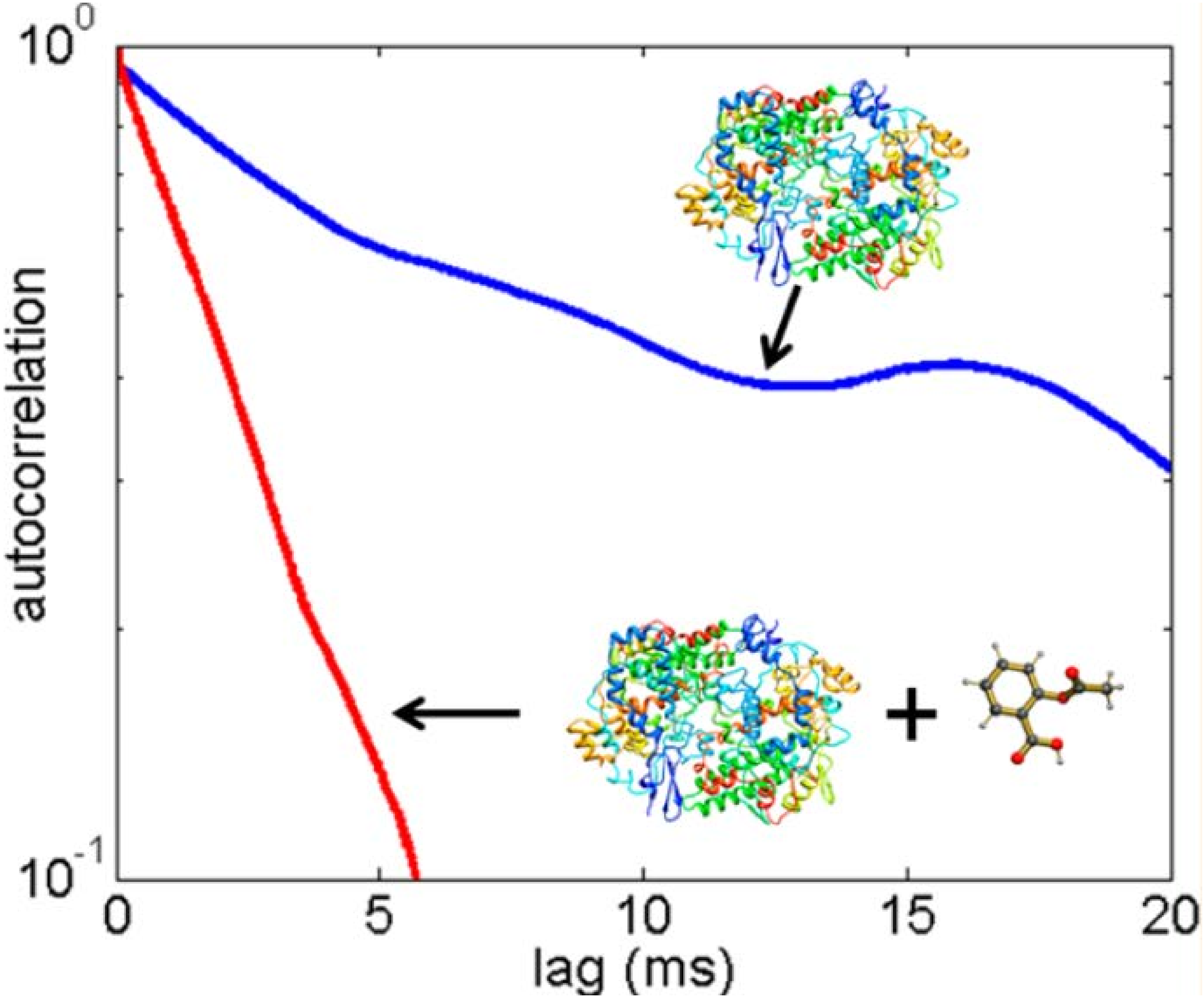
Protein – Small Molecule Interactions

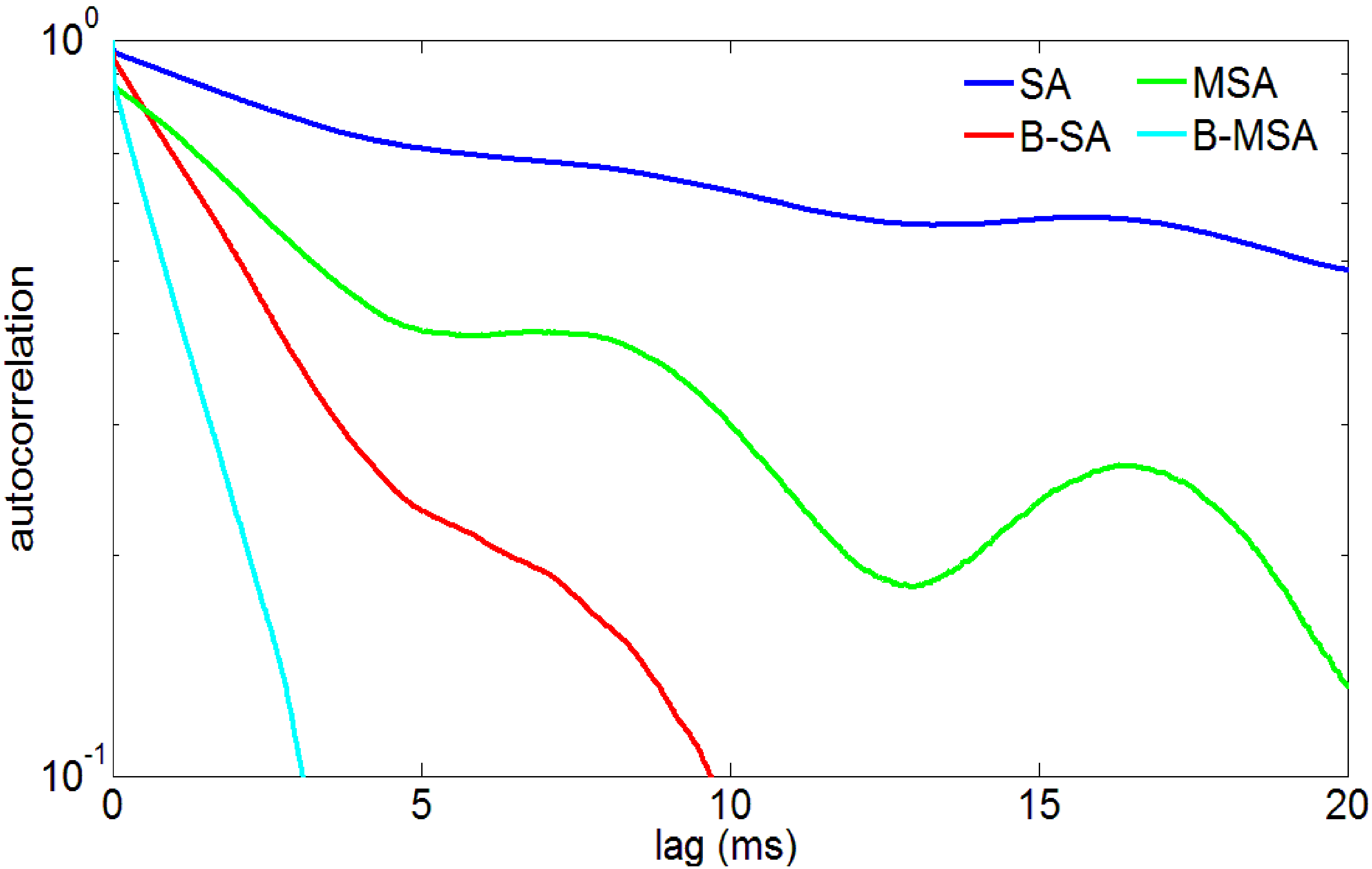






Protein-Small Molecule Binding

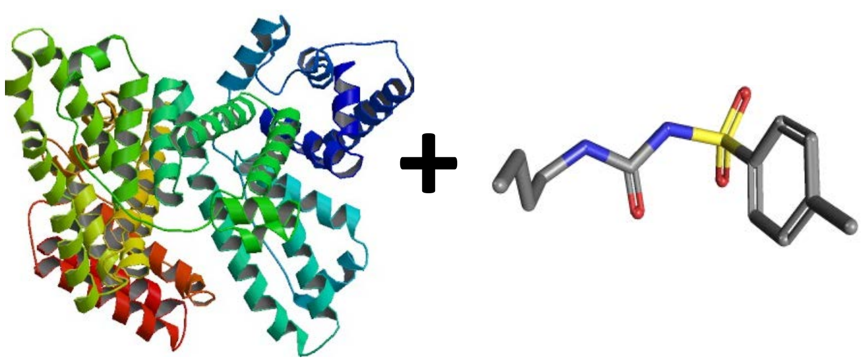


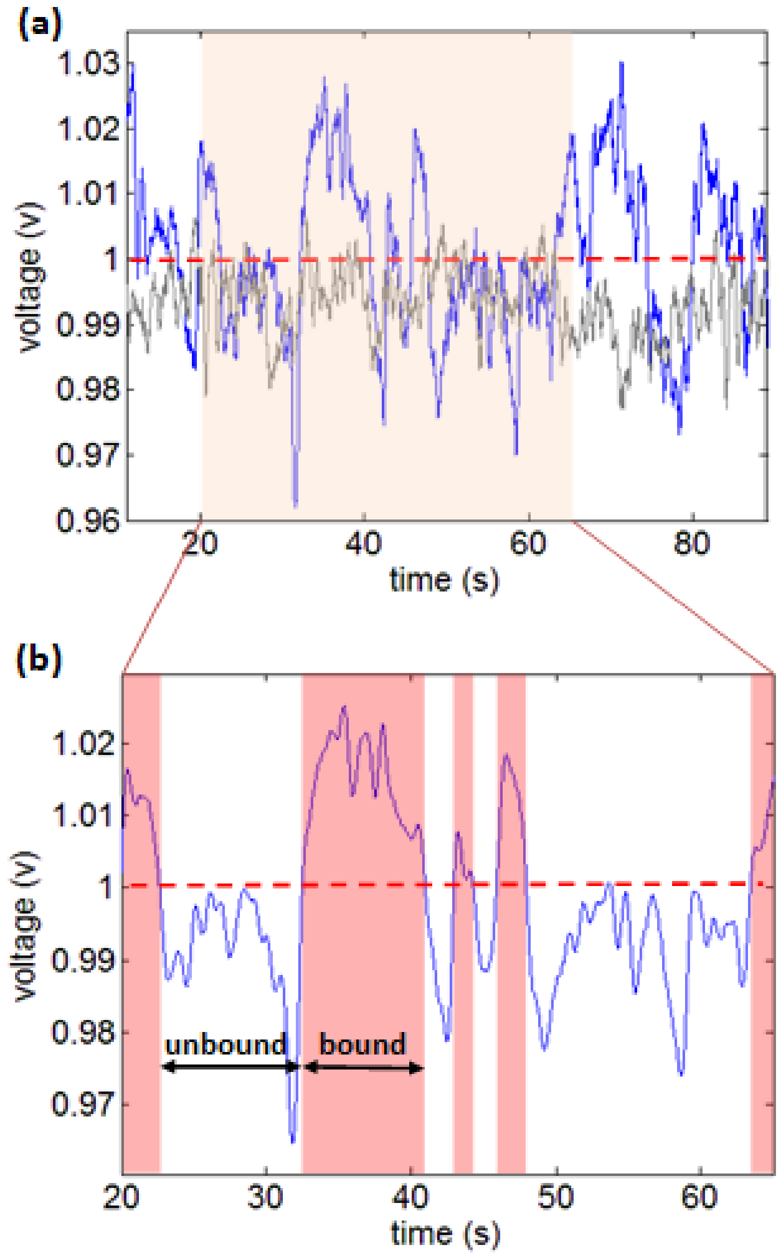


A. A. Al Balushi, R. Gordon, " Label-Free Free-Solution Single-Molecule Protein.Small Molecule Interaction Observed by Double-Nanohole Plasmonic Trapping," *ACS Photonics,* 1(5), 389-393 (2014).

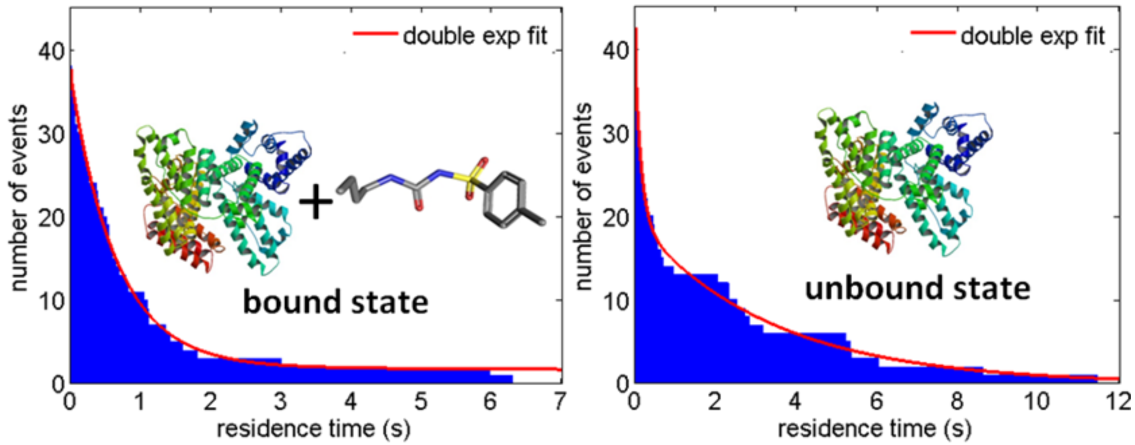
HSA Binding Kinetics

$$P + L \underset{k_d}{\overset{k_a}{\rightleftharpoons}} PL$$

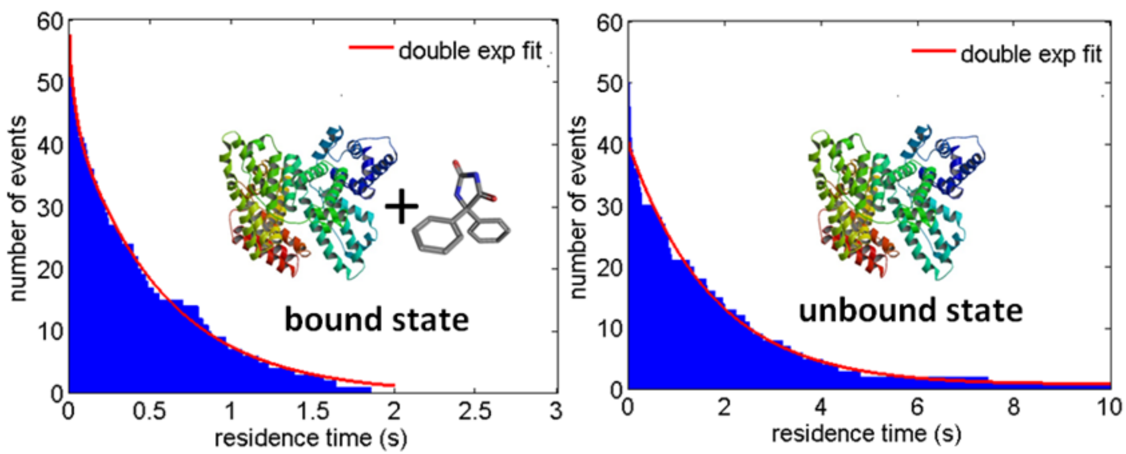




HSA binding kinetics



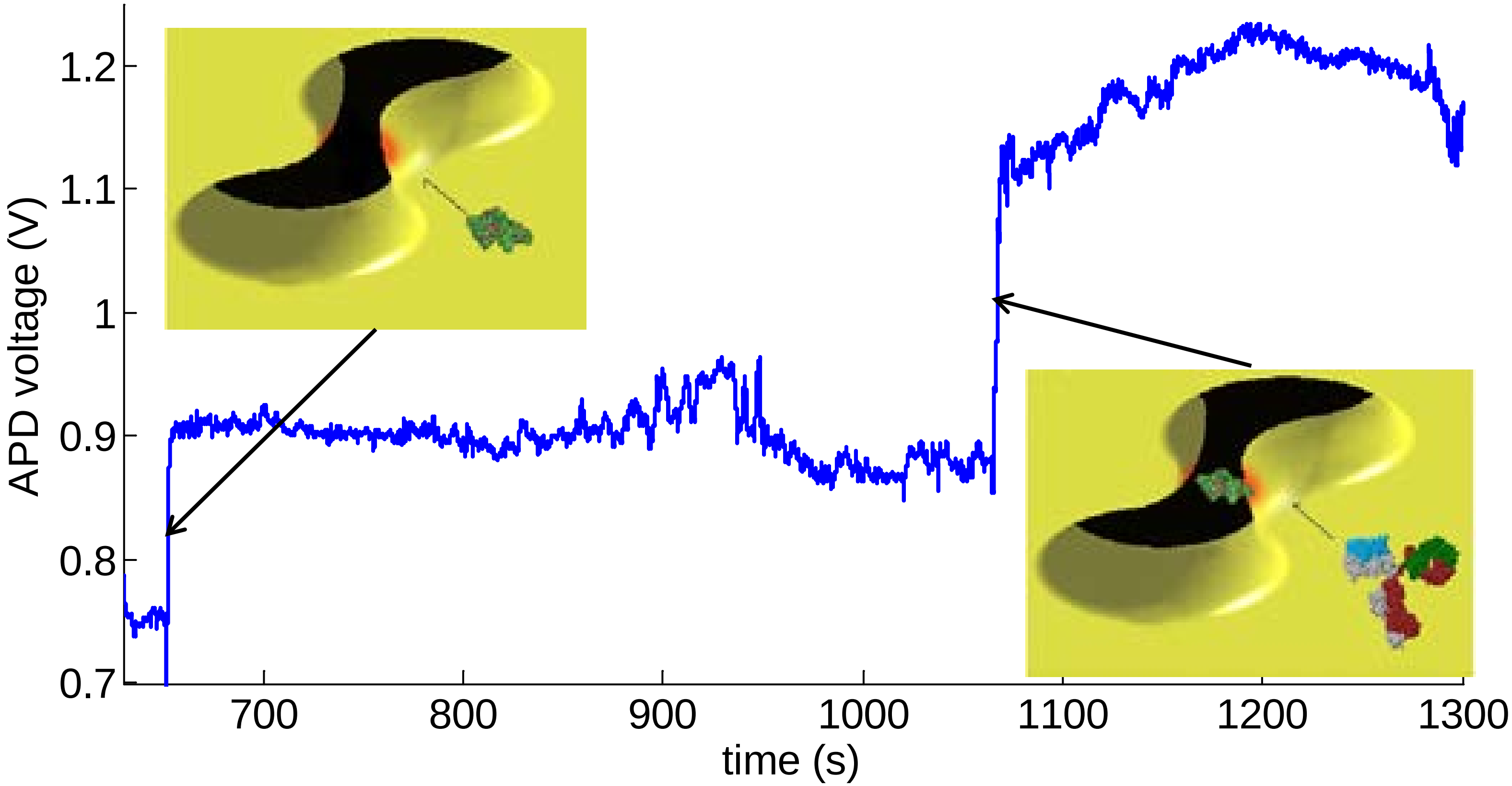
Tolubutamide: 94.7 µM
Literature 71 - 111 µM



Phenytoin: 13 µM
Literature 4.5-31 µM

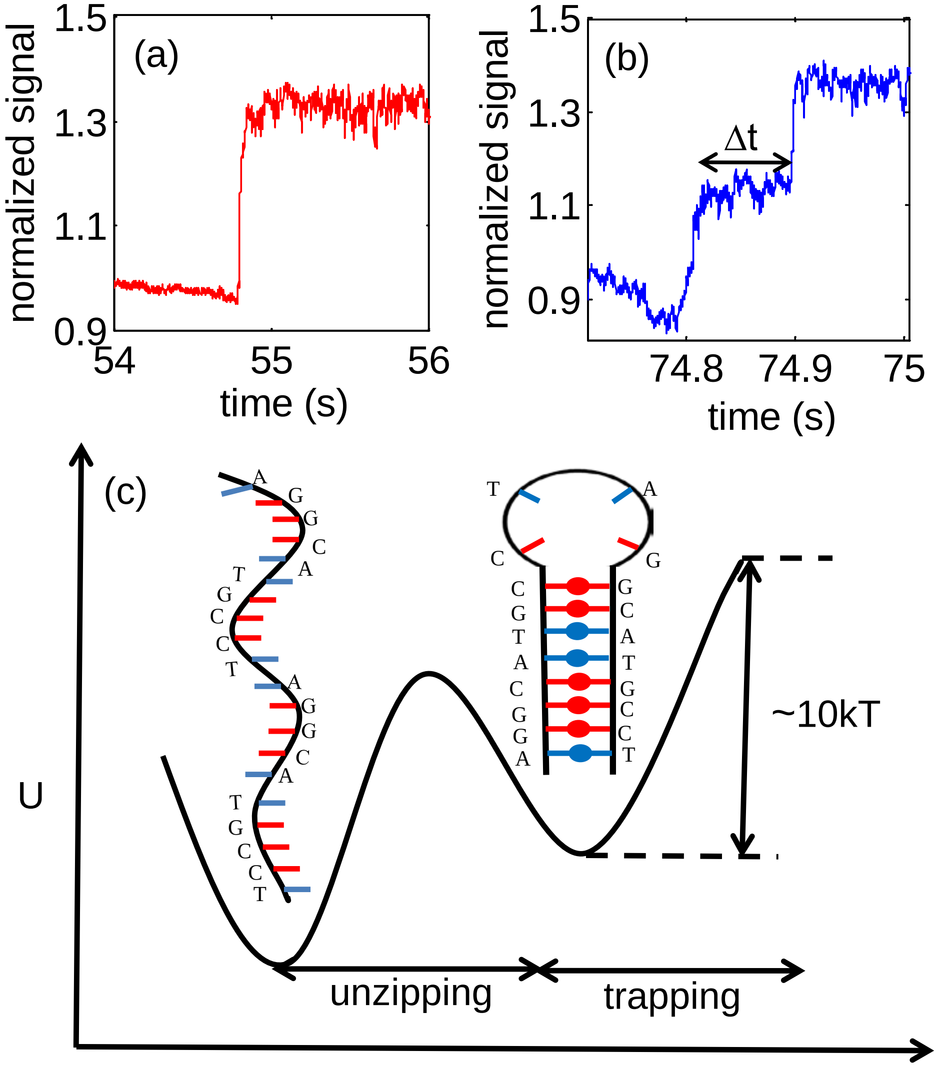
A. A. Al Balushi, R. Gordon, "A Label-Free Untethered Approach to Single-Molecule Protein Binding Kinetics," Nano Letters 14 (10), 5787-5791 (2014).

Protein-Antibody Binding

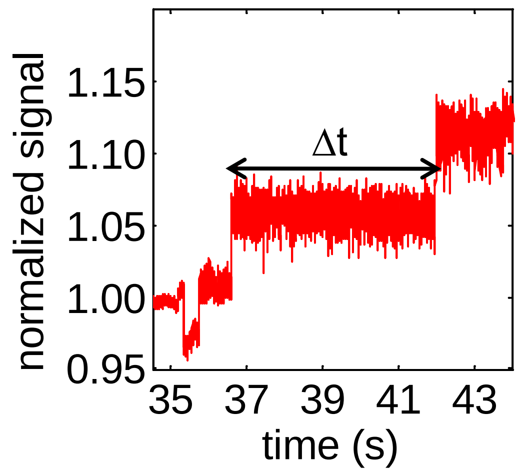


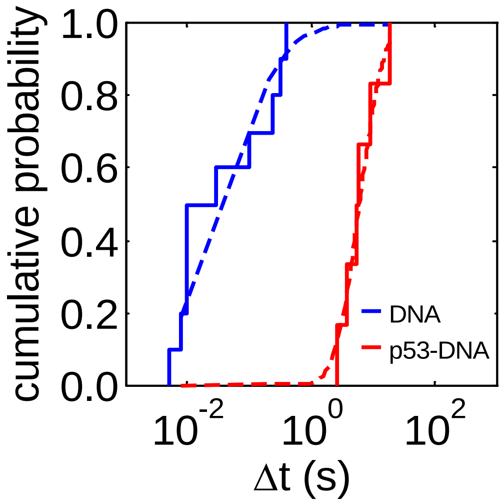
A. Zehtabi-Oskuie, H. Jiang, B. Cyr, D. Rennehan, A. Al-Balushi, R Gordon, "Double nanohole optical trapping: Dynamics and protein-antibody co-trapping," *Lab Chip,* 13, 2563-2568 (2013).

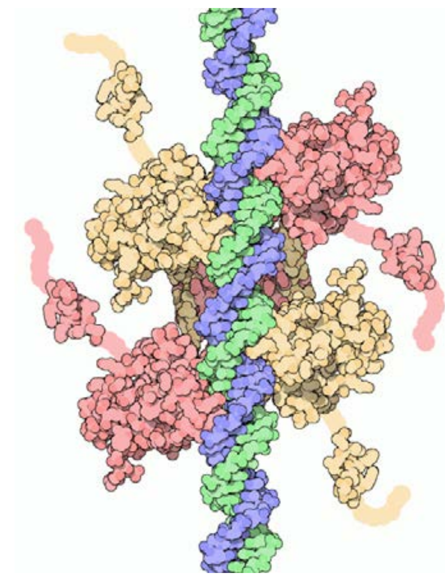
Unzipping 10 bp DNA

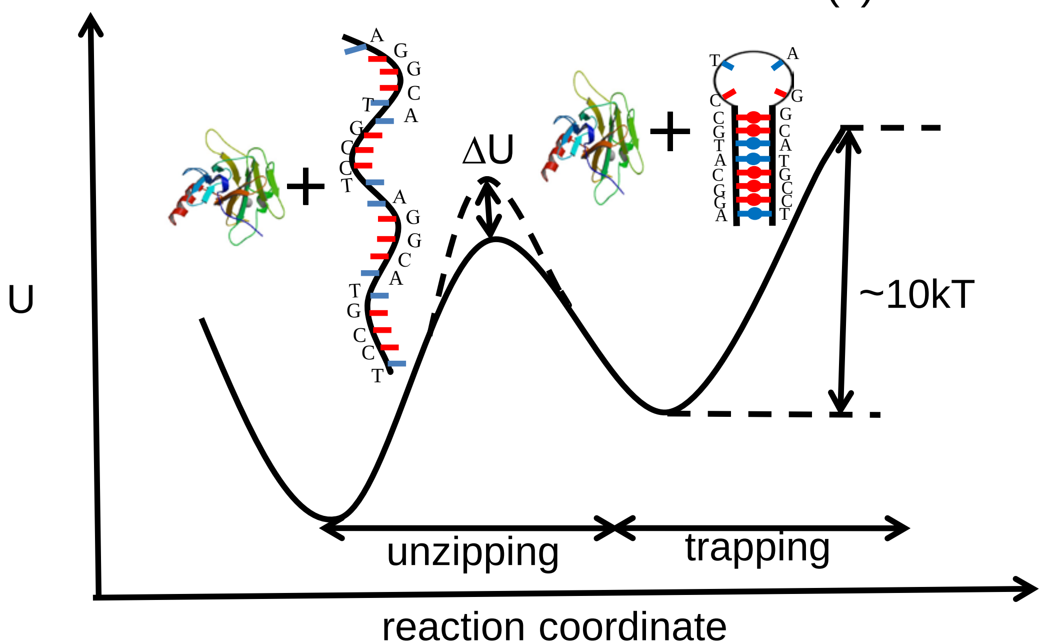


Protein DNA interactions





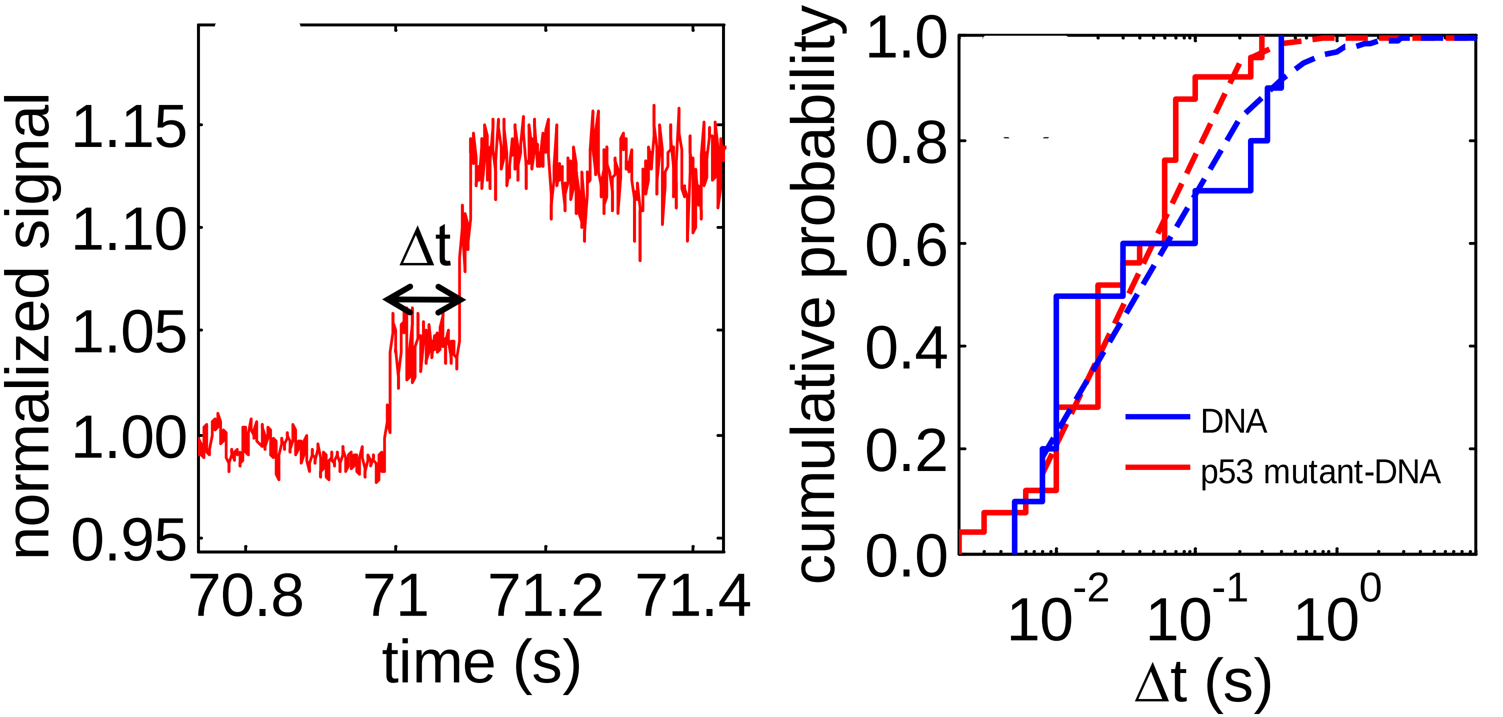




$$\Delta U = -kT \ln \frac{t_{p53}}{t_{DNA}}$$

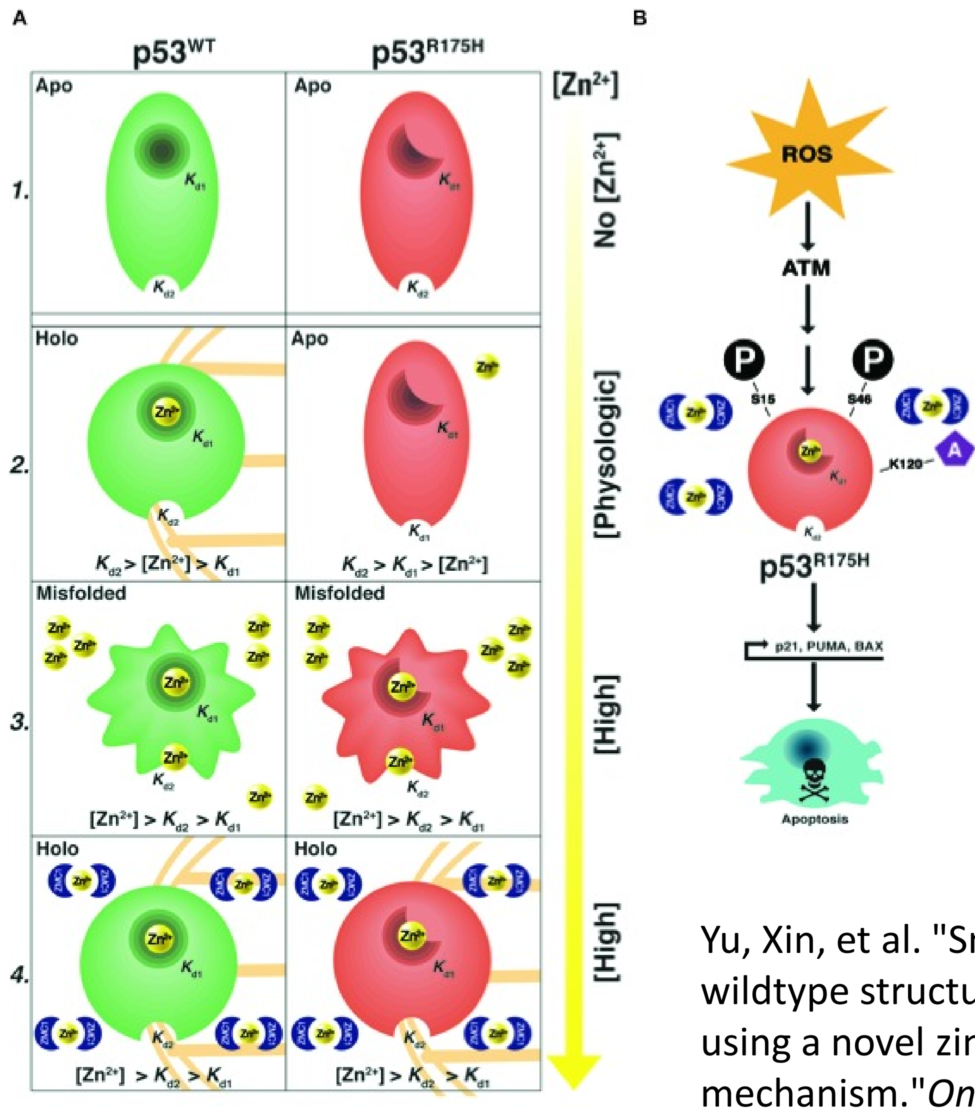
$$\Delta U = -2 \times 10^{-20}\,\mathrm{J}$$

Mutant p53 ineffective

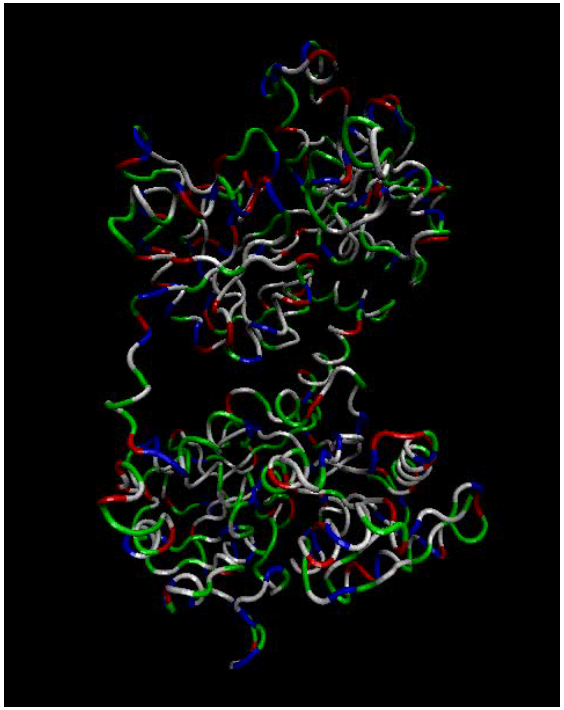


A. Kotnala, R. Gordon, "Double nanohole optical tweezers visualize protein p53 suppressing unzipping of single DNA-hairpins," *Biomedical Optics Express,* 5(6), 1886-1894 (2014).

p53 misfolding



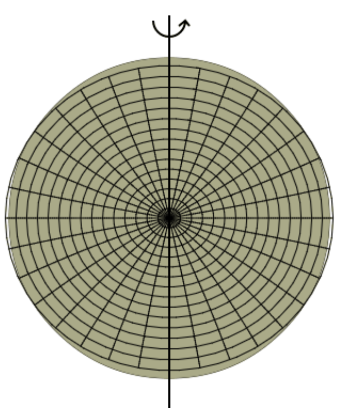
Yu, Xin, et al. "Small molecule restoration of wildtype structure and function of mutant p53 using a novel zinc-metallochaperone based mechanism."*Oncotarget* 5.19 (2014): 8879.

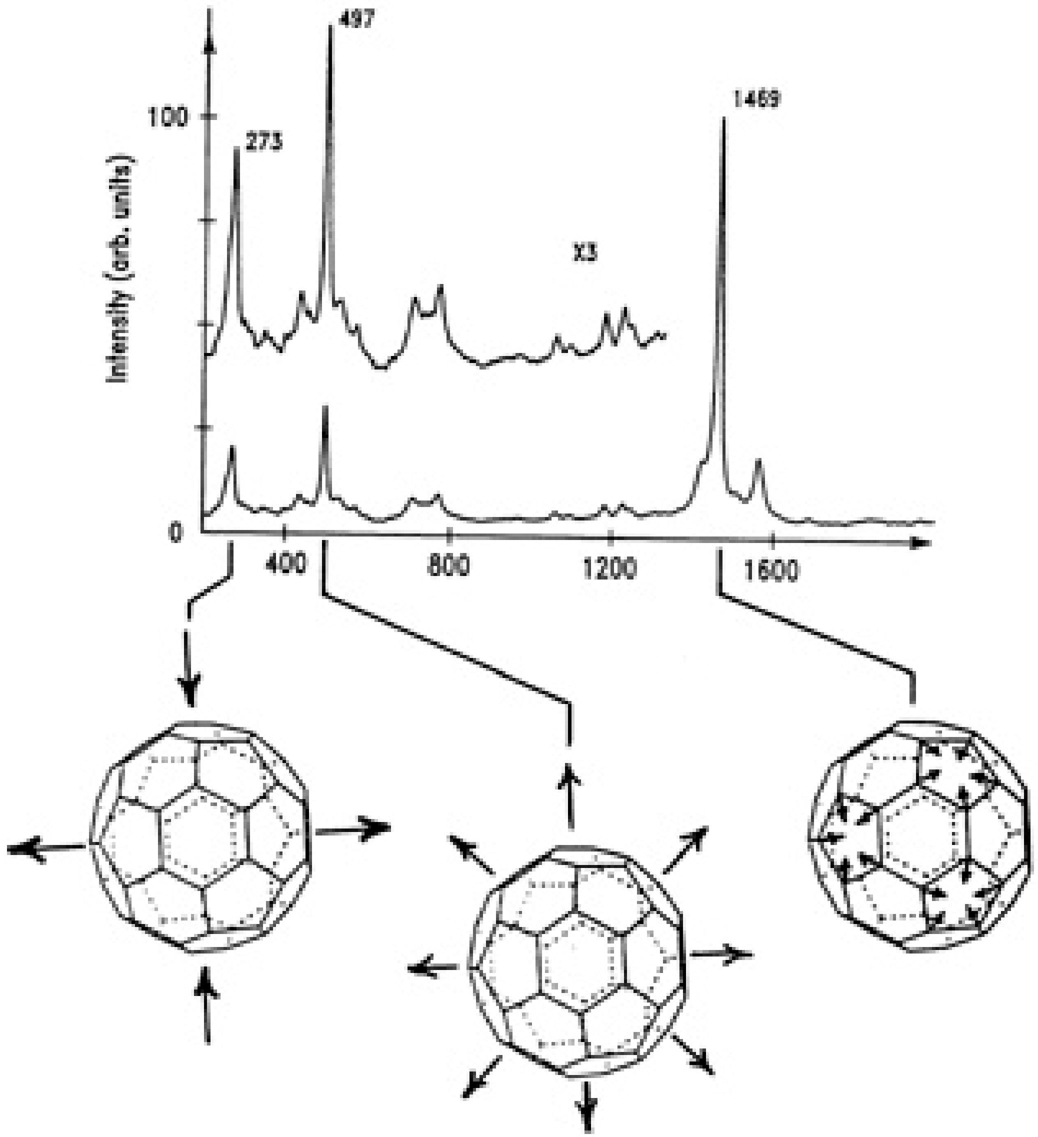


EXTRAORDINARY ACOUSTIC RAMAN SCATTERING (EARS)

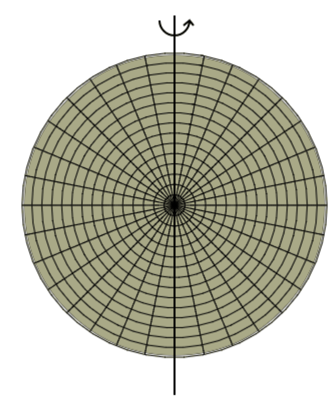
Nanoparticle Vibrational Modes: C60

8.18 THz

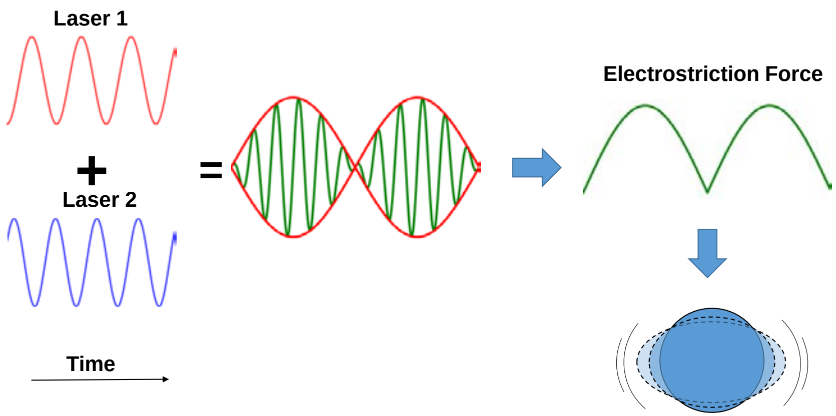


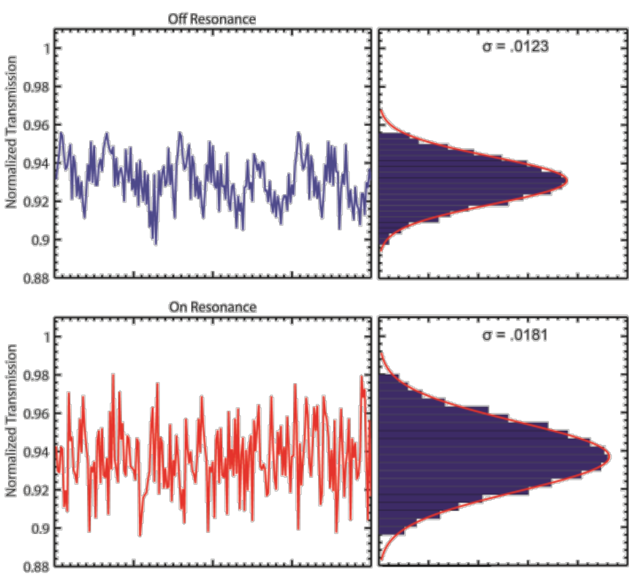


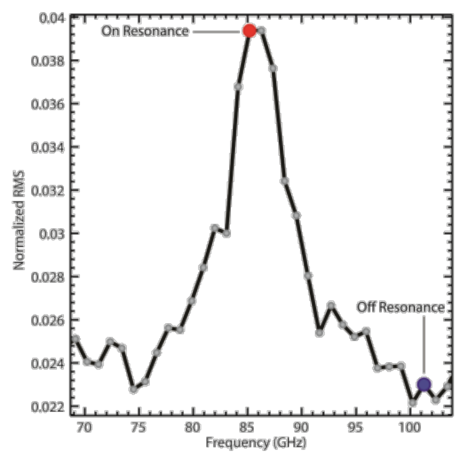
14.89 THz

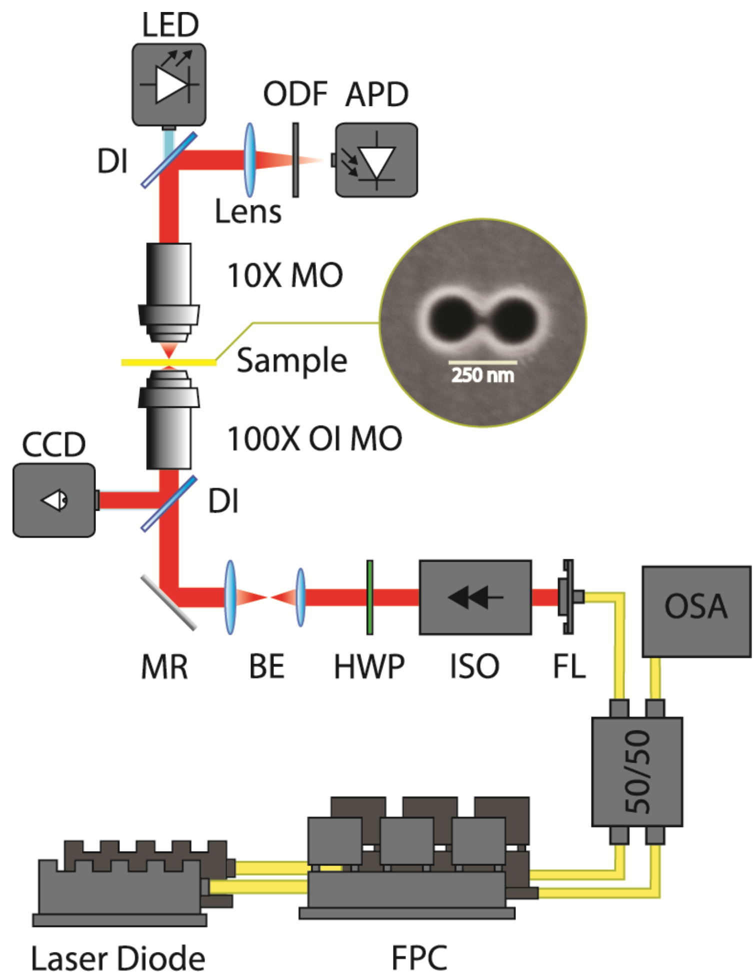


Extraordinary Acoustic Raman Scattering (EARS)

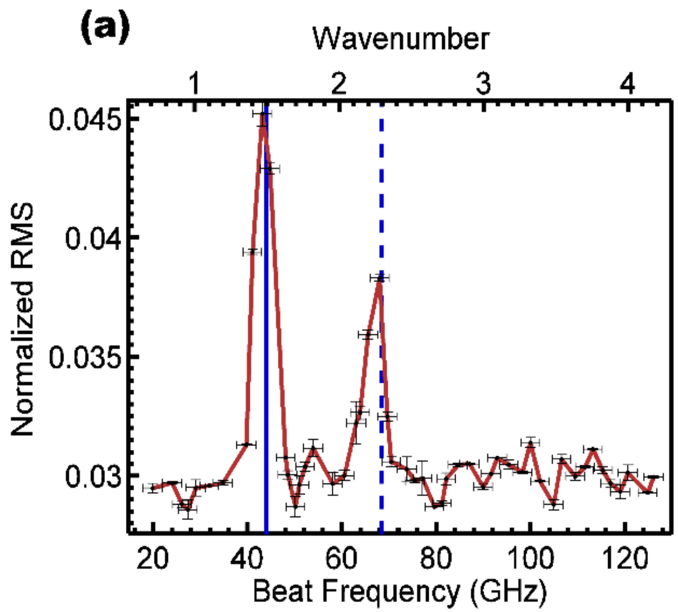


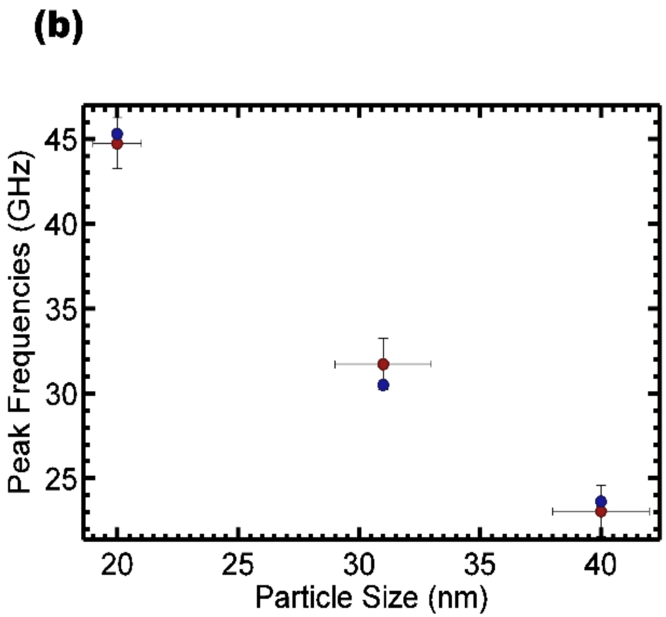


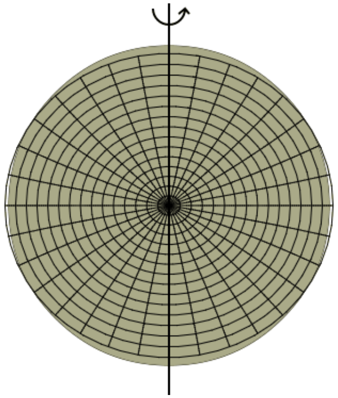


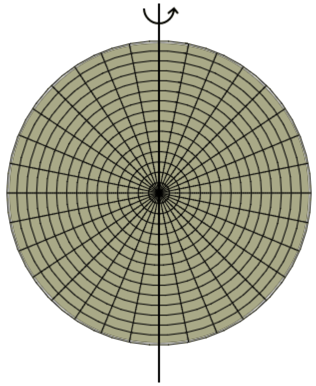


Acoustic Modes of Nanospheres



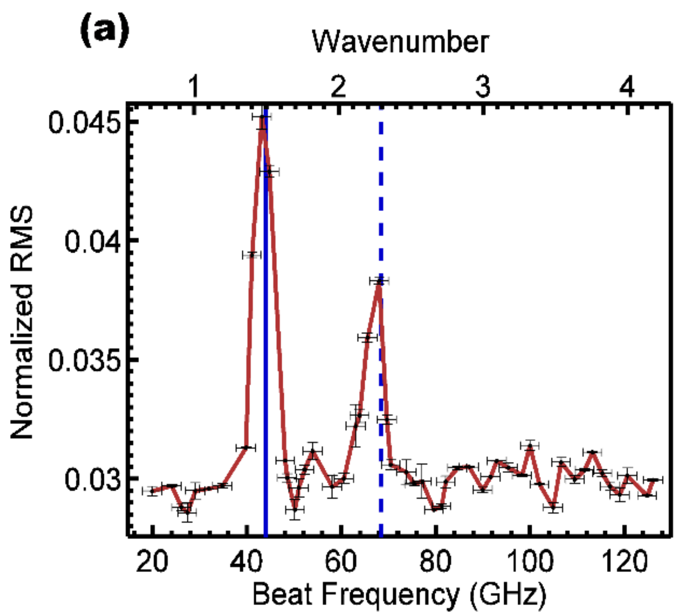


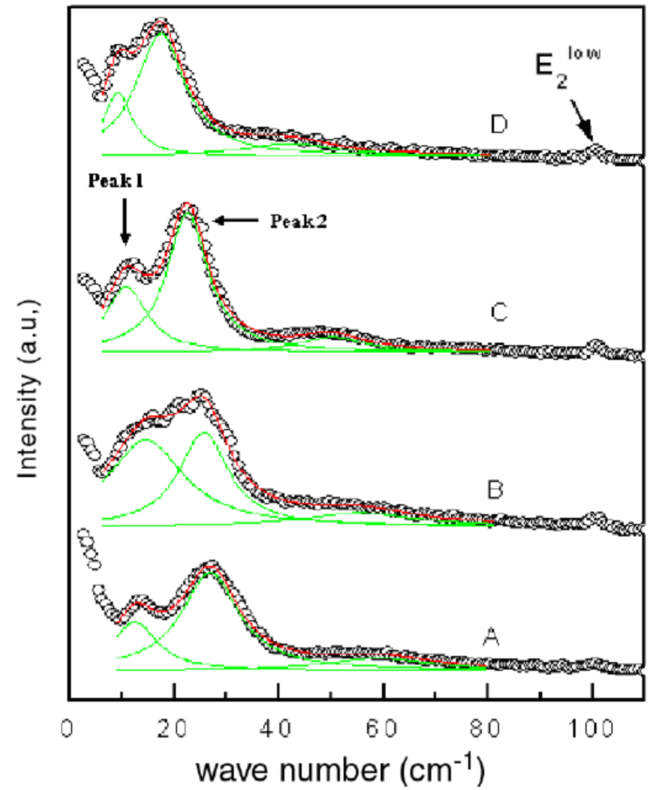




S. Wheaton, R. M. Gelfand, R. Gordon, "Probing the Raman-Active Acoustic Vibrations of Nanoparticles with Extraordinary Spectral Resolution" Nature Photonics Photonics 9, 68-72 (2015).

Acoustic Modes of Nanospheres

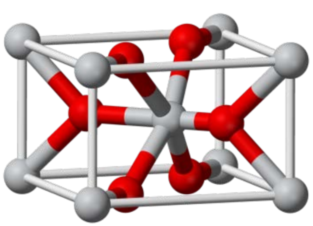


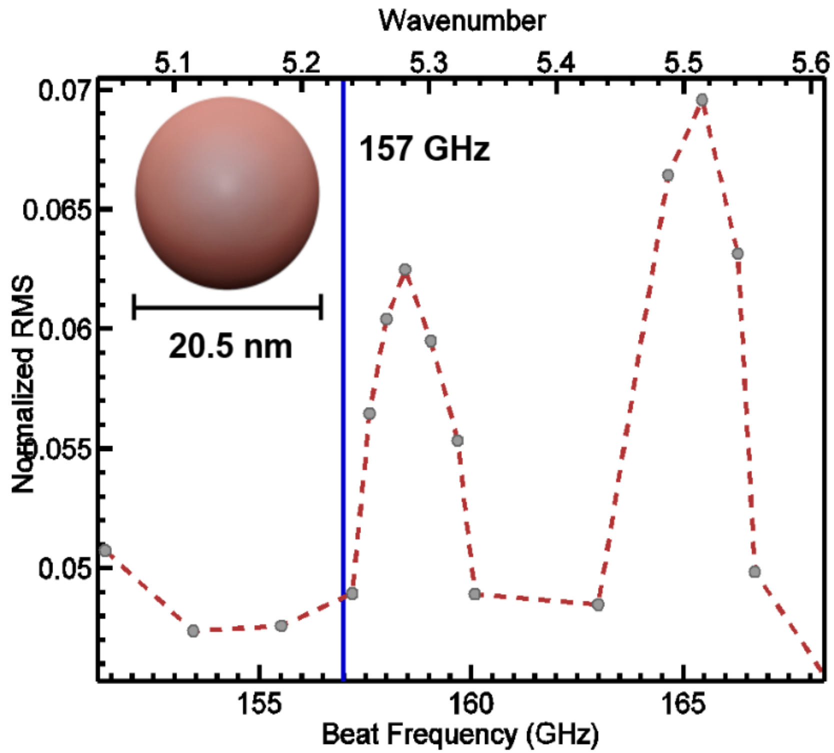


Physical Review Letters 97, 085502 (2006).

Conventional Raman suffers from ensemble averaging, zero loss (Rayleigh) line and instrument resolution. Micro-Brillouin can probe down to about 200 nm single particles.

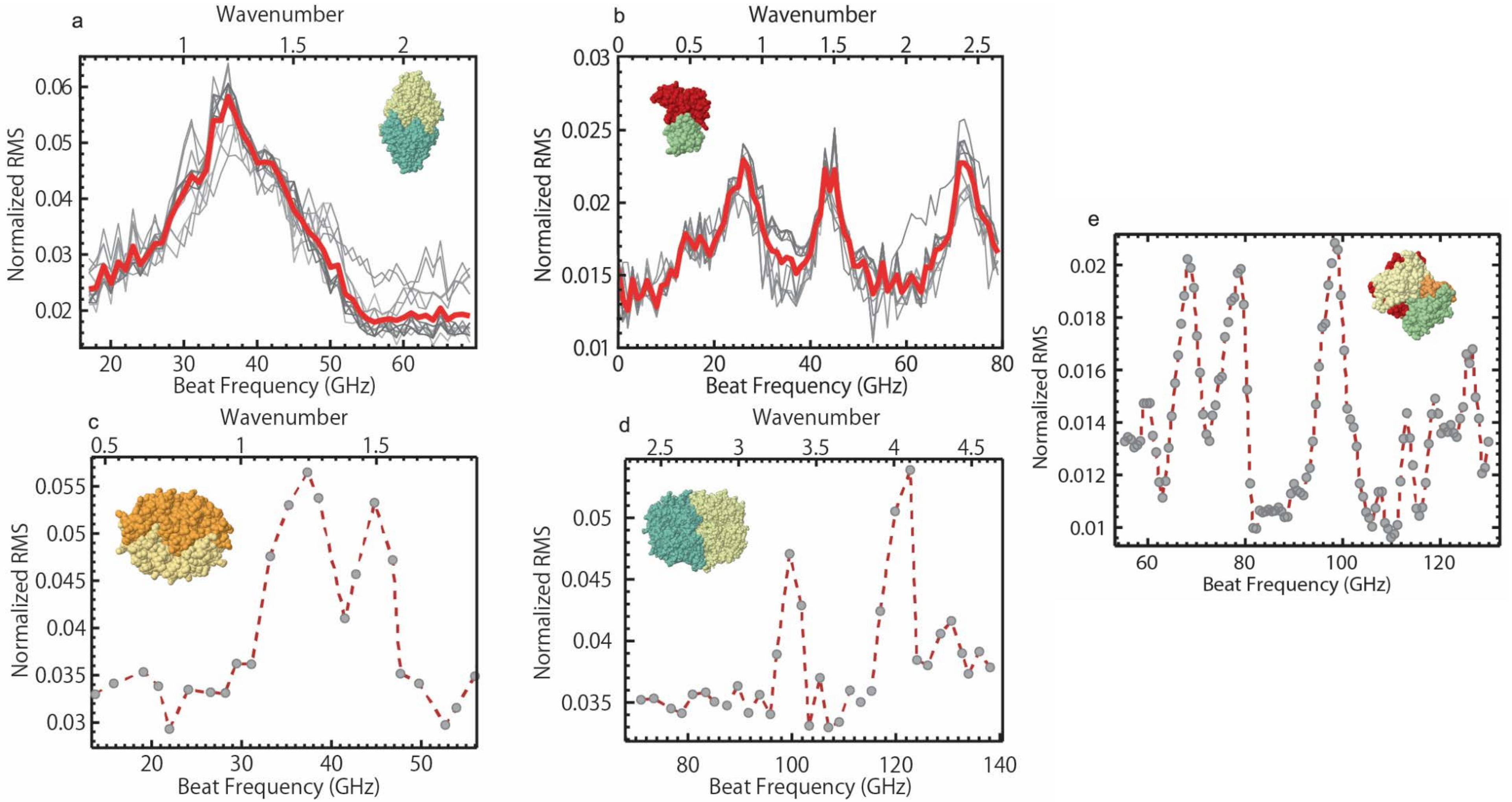
Probing Material Anisotropy



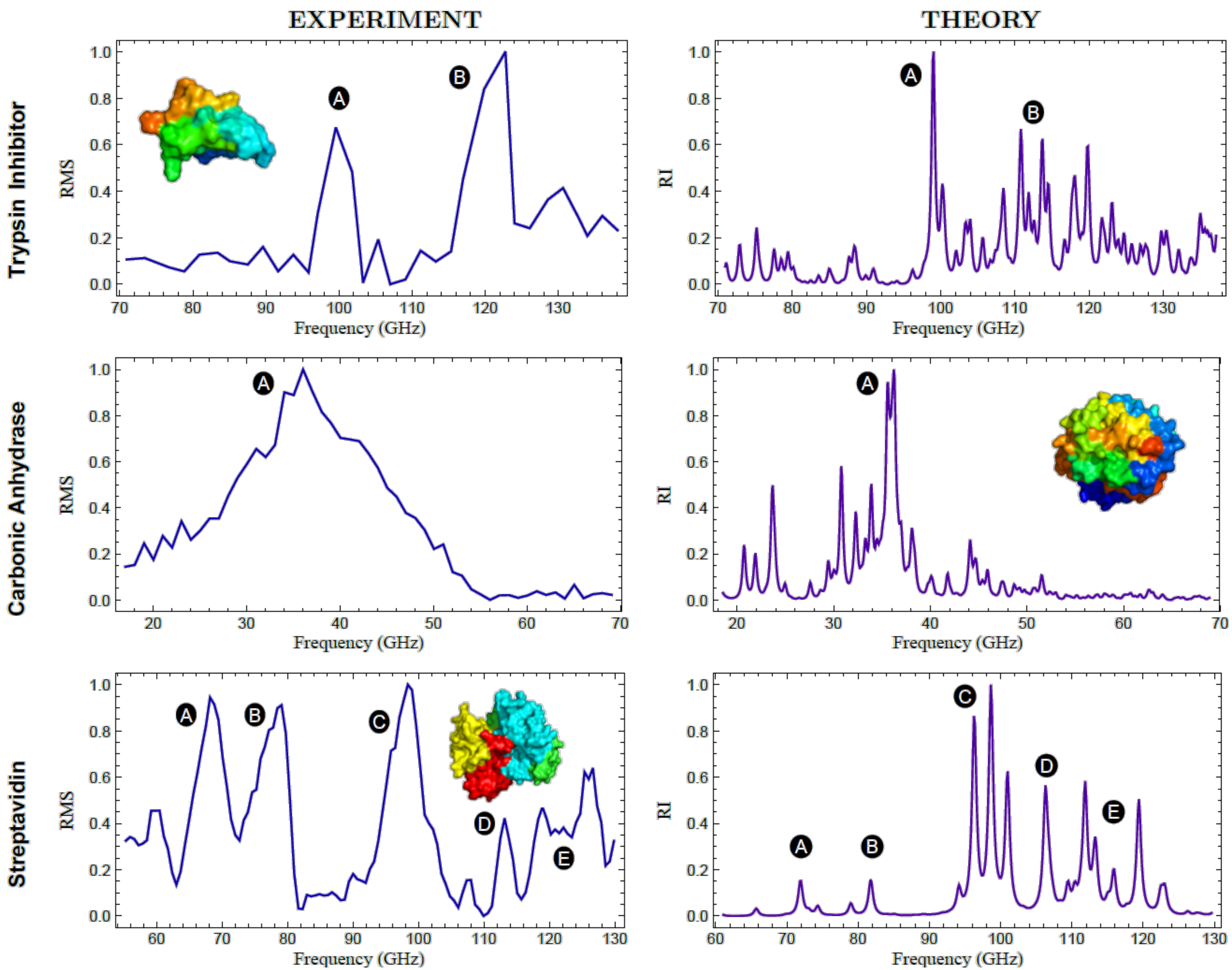


Titania anatase

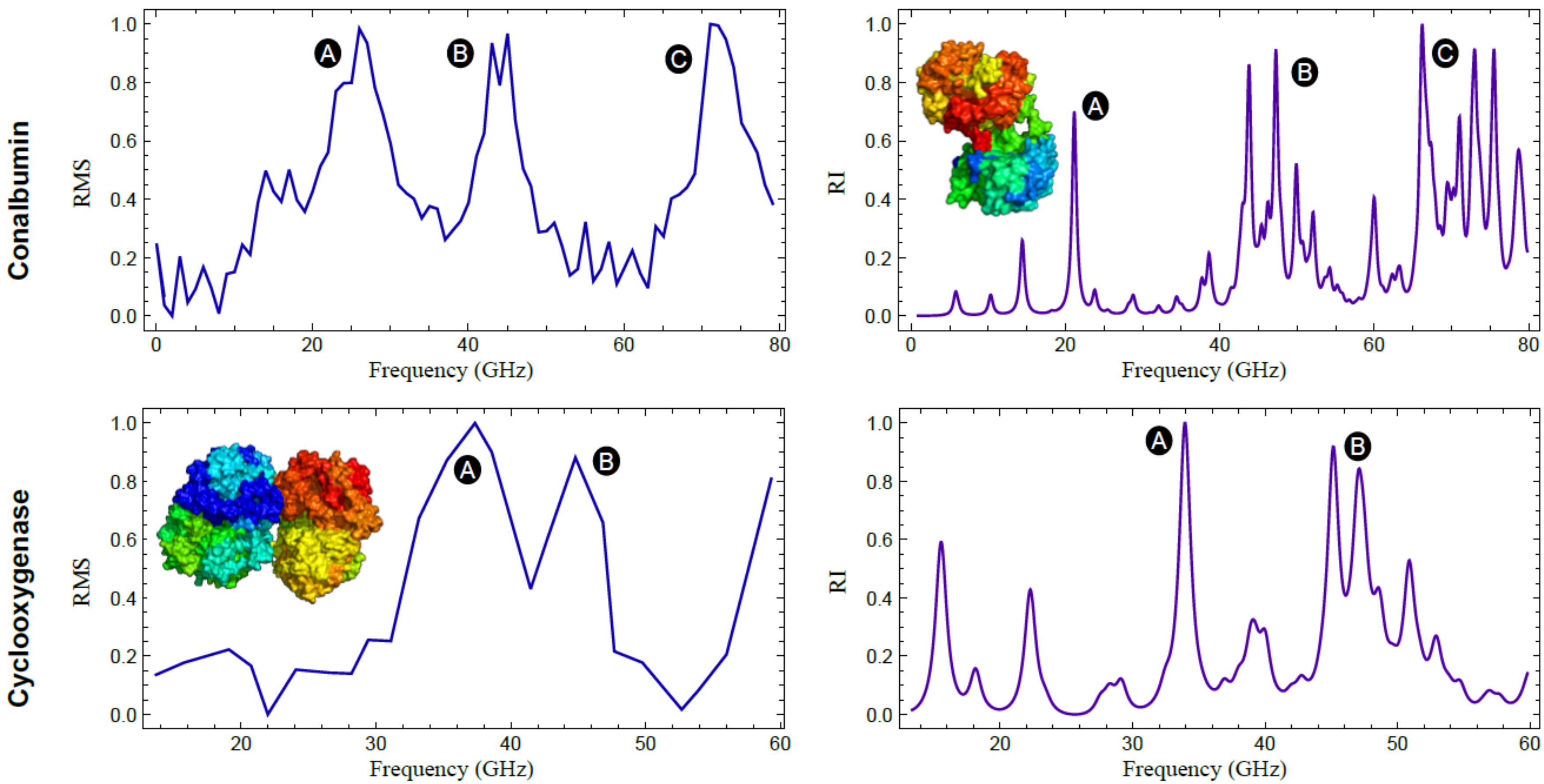
Acoustic Modes of Proteins



Raman Analysis

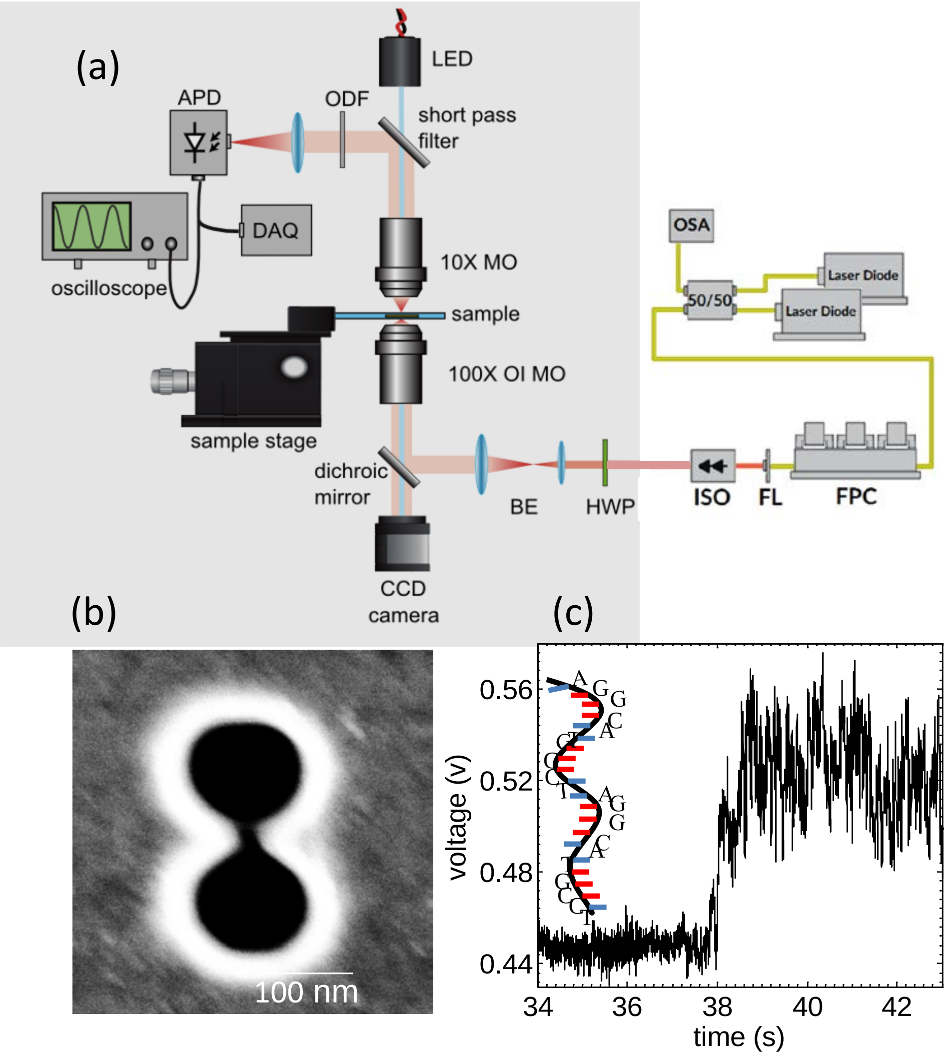


Raman theory cont.

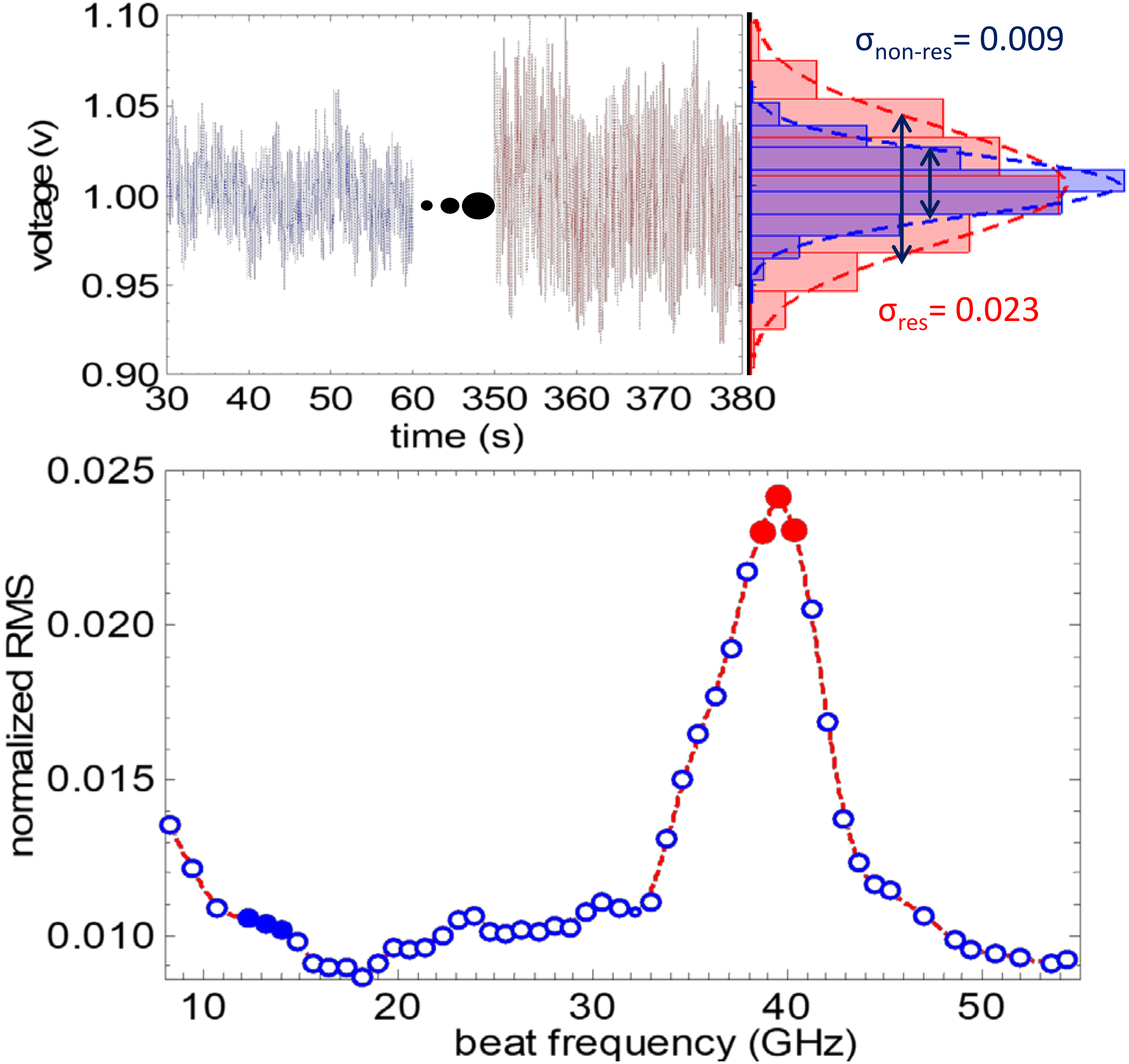


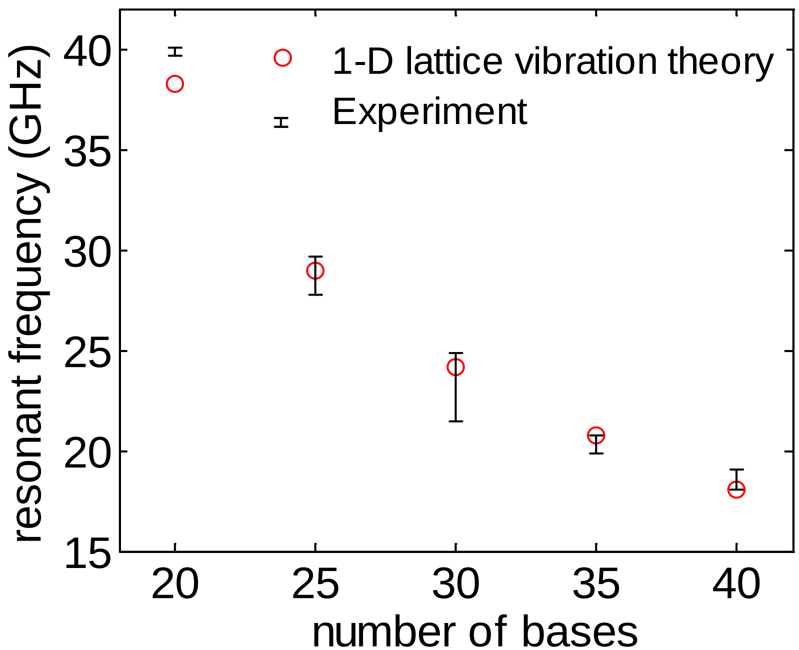
T. DeWolf, R. Gordon, "Theory of Acoustic Raman Modes in Proteins," *Physical Review Letters* 117, 138101 (2016).

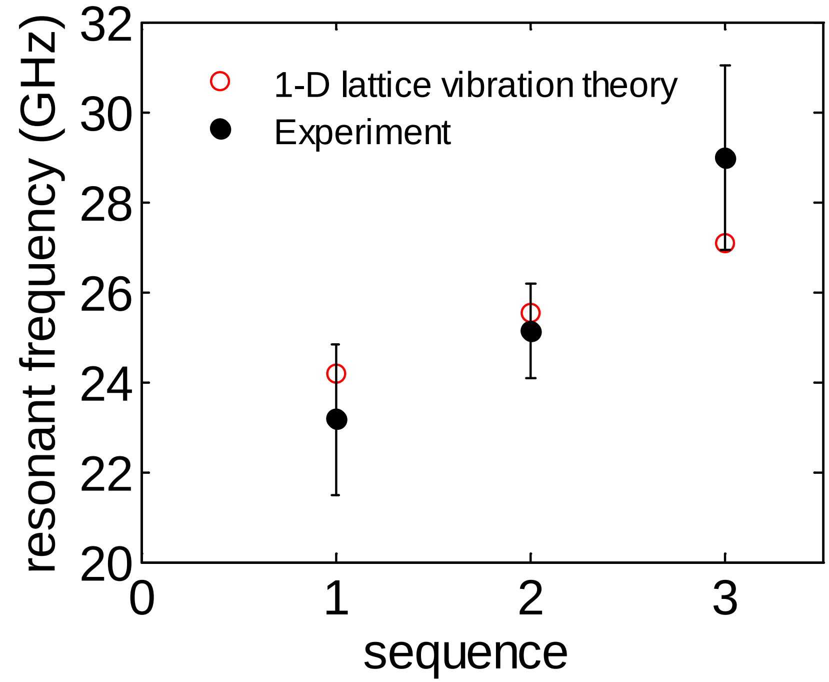
Acoustic Modes of ssDNA



Acoustic Modes of ssDNA







Seq 1: M=144.5Da, Seq 2: M=129.8Da, Seq 3: M=115.4Da

$$\omega \approx \frac{\pi}{N_B}\sqrt{\frac{\kappa}{M}}$$

ω : resonant vibrational frequency

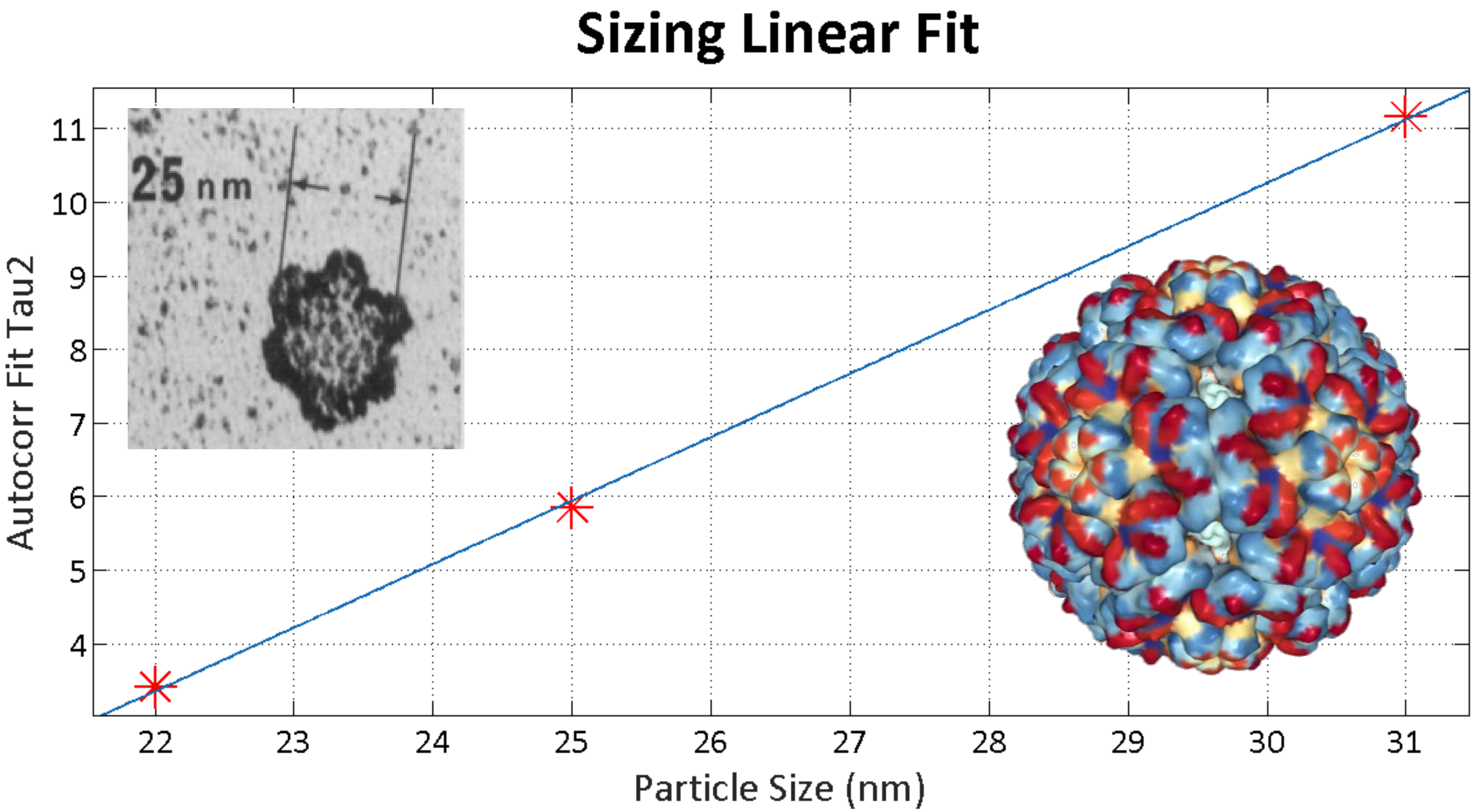
N_B : number of bases

κ : spring constant of DNA strand

M : weighted average mass of DNA strand

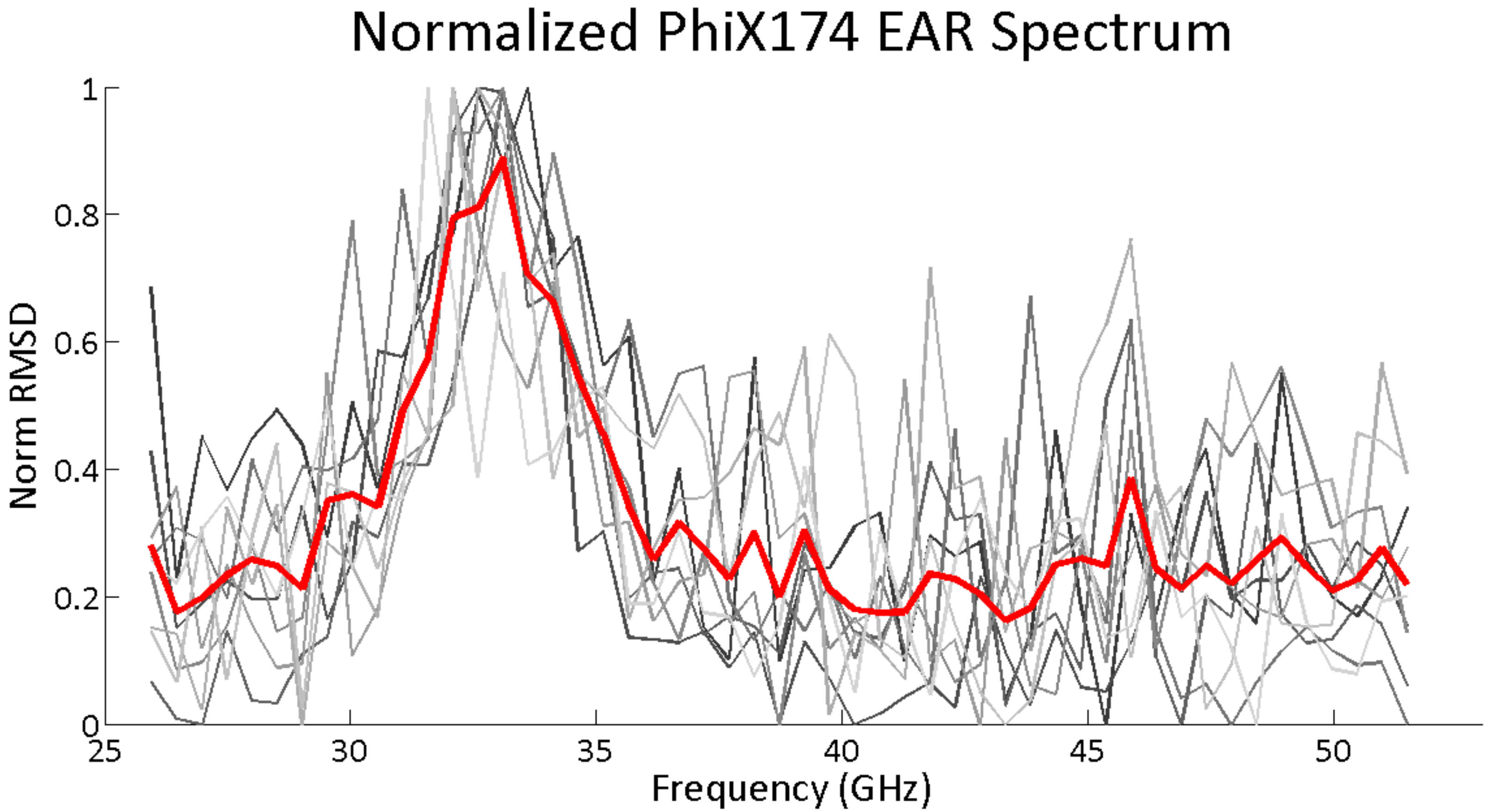
A. Kotnala, S. Wheaton, R. Gordon, "Playing the notes of DNA with light: extremely high frequency nanomechanical oscillations," *Nanoscale,* 7, 2295-2300 (2015).

Sizing Viruses (PhiX174)

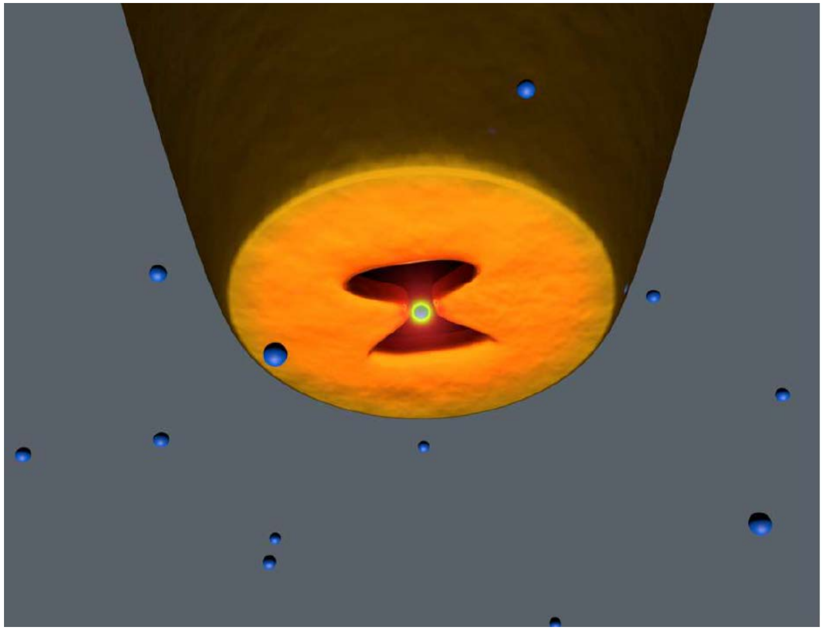


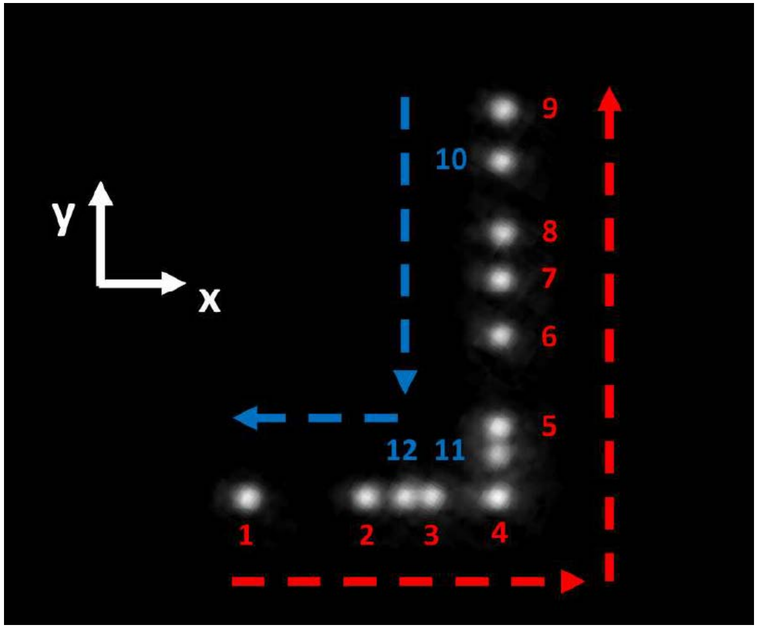
w/ Jeff Burkhartsmeyer and Yanhong Wang (visiting reseachers)

ΦX174 EAR Spectrum



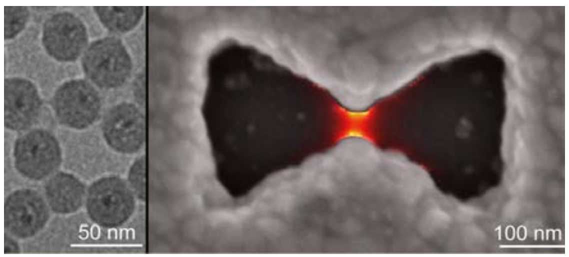
Optical Nanopipette (Quidant group)

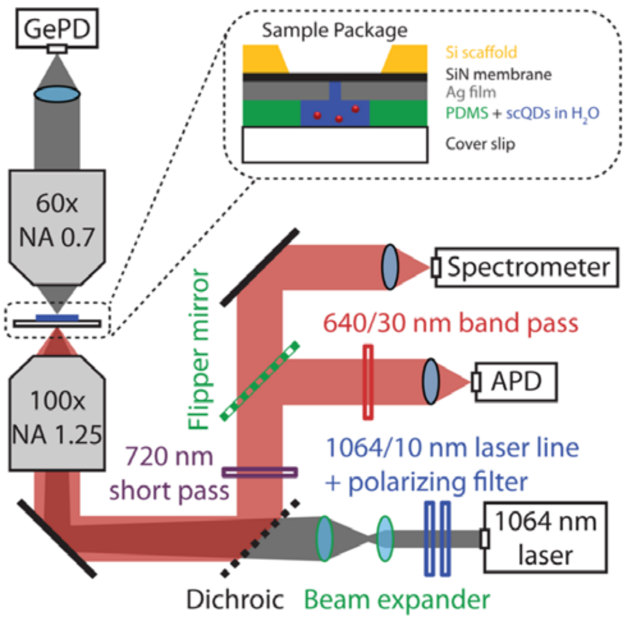


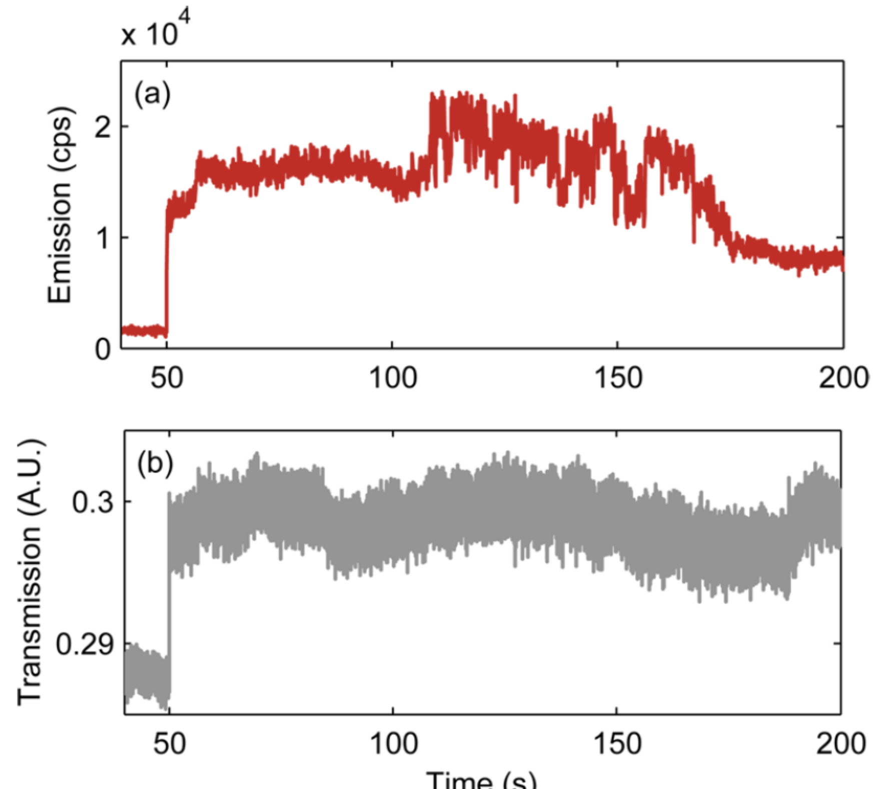


Berthelot, J., Aćimović, S. S., Juan, M. L., Kreuzer, M. P., Renger, J., & Quidant, R. (2014). Three-dimensional manipulation with scanning near-field optical nanotweezers. *Nature Nanotechnology*, *9*(4), 295-299.

Quantum Dots – Two Photon Excitation (Loncar/Bawendi groups)

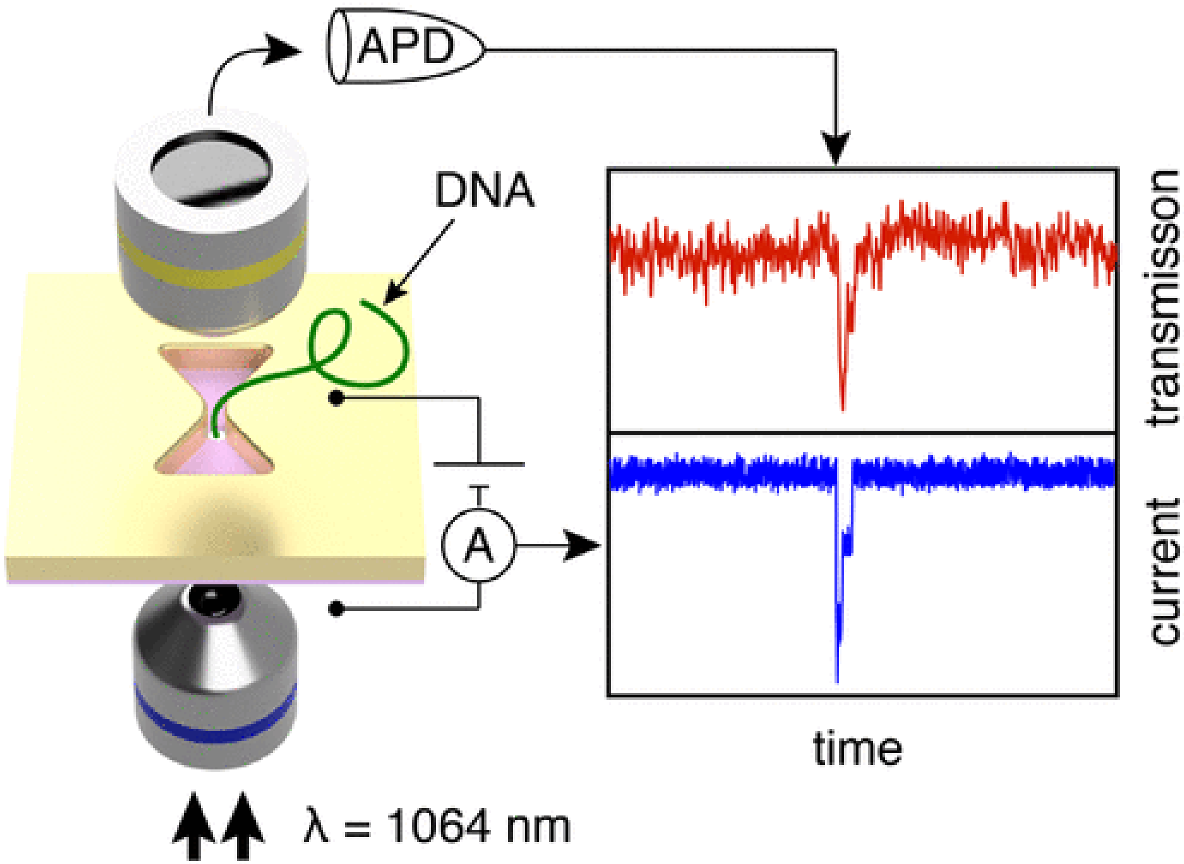






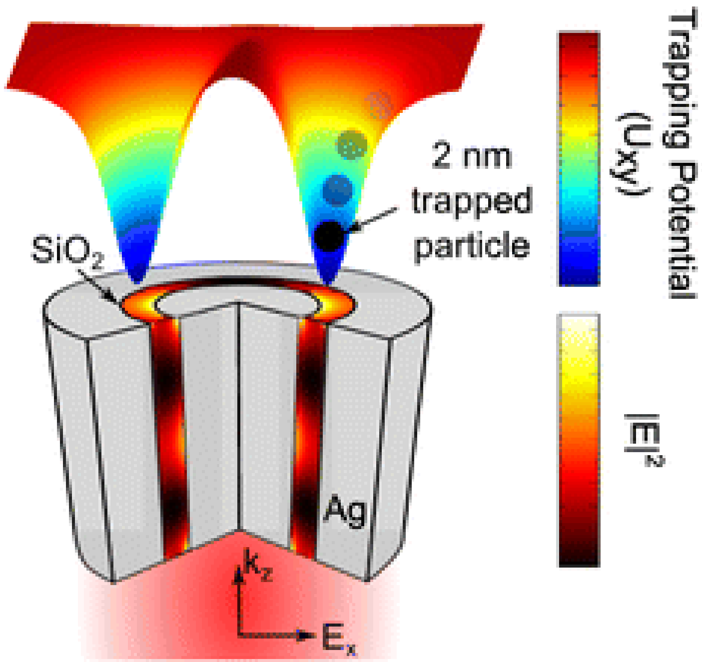
Jensen, R. A., Huang, I. C., Chen, O., Choy, J. T., Bischof, T. S., Lončar, M., & Bawendi, M. G. (2016). Optical trapping and two-photon excitation of colloidal quantum dots using bowtie apertures. *ACS Photonics*, *3*(3), 423-427.

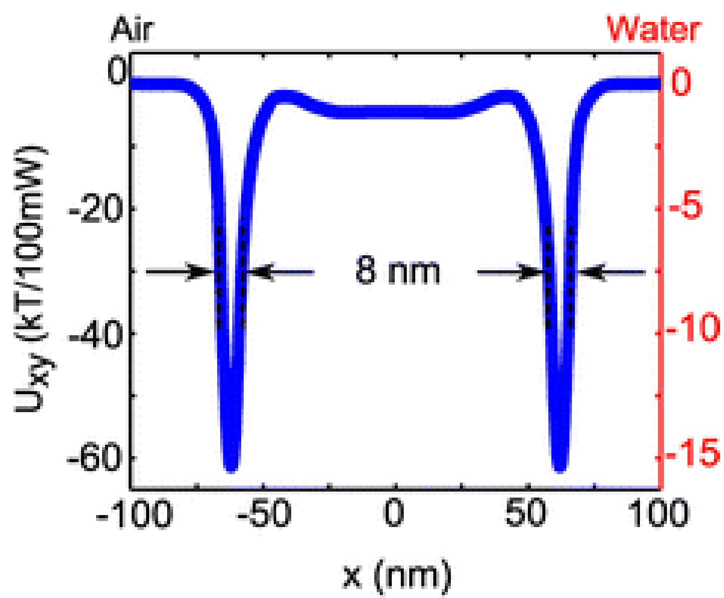
Nanopore Translocation of DNA Measured Optically (Dekker/Kuipers groups)



Label-Free Optical Detection of DNA Translocations through Plasmonic Nanopores
Daniel V. Verschueren, Sergii Pud, Xin Shi, Lorenzo De Angelis, L. Kuipers, and Cees Dekker
ACS Nano **Article ASAP**
DOI: 10.1021/acsnano.8b06758

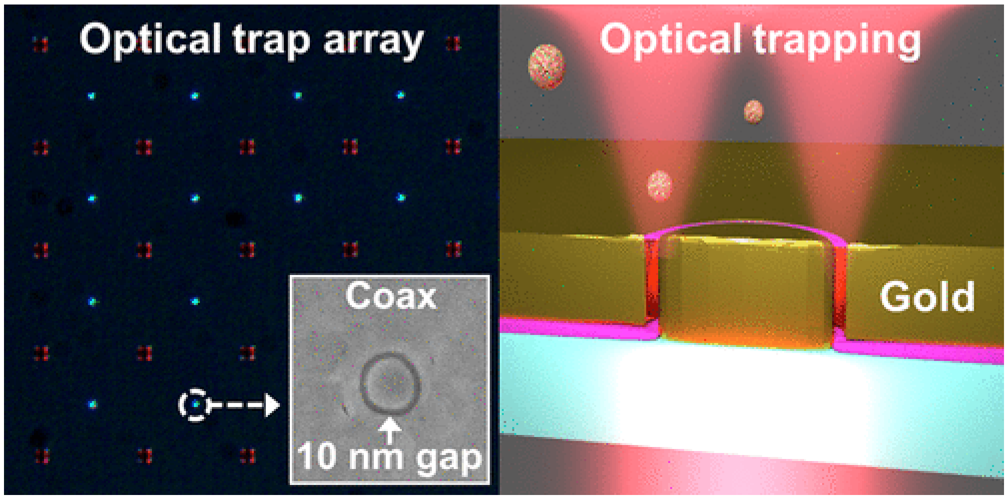
Coaxial Trapping (Dionne group)

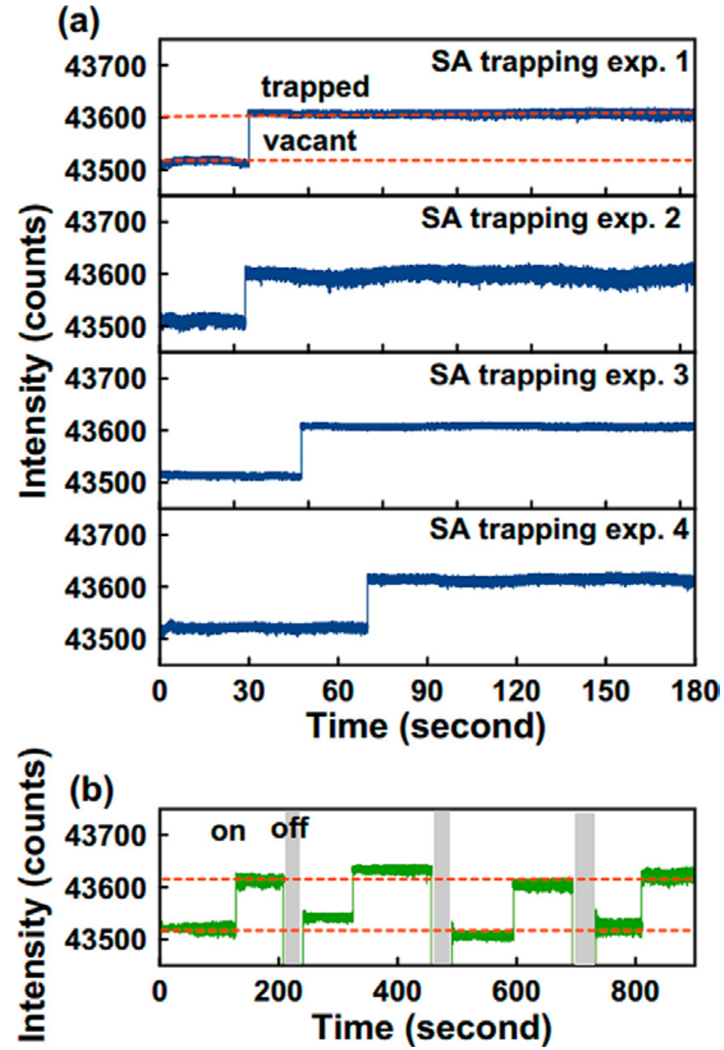




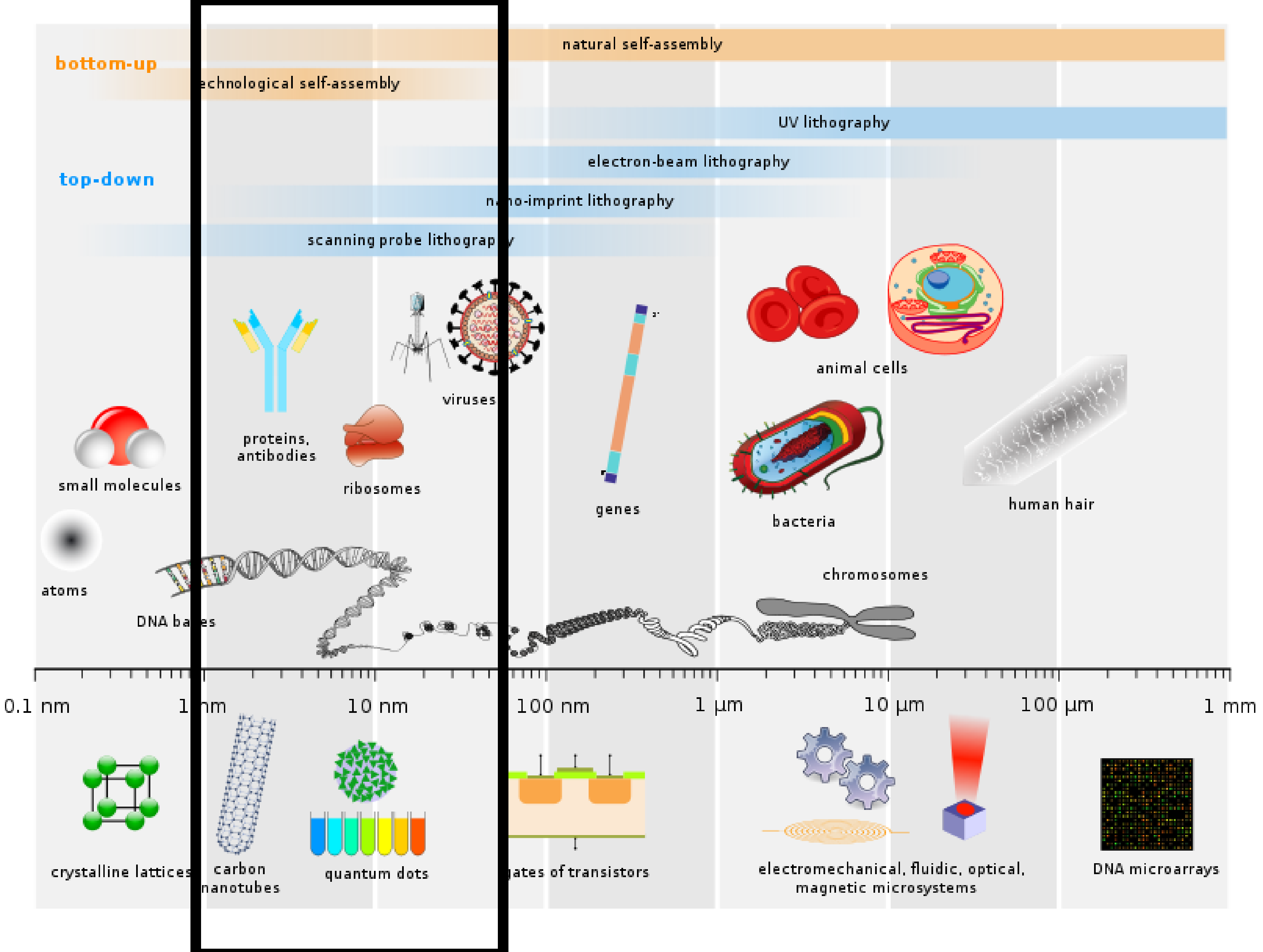
Saleh, A. A., & Dionne, J. A. (2012). Toward efficient optical trapping of sub-10-nm particles with coaxial plasmonic apertures. *Nano Letters*, *12*(11), 5581-5586.

Reliable Mass Fabrication (w/ Oh group)

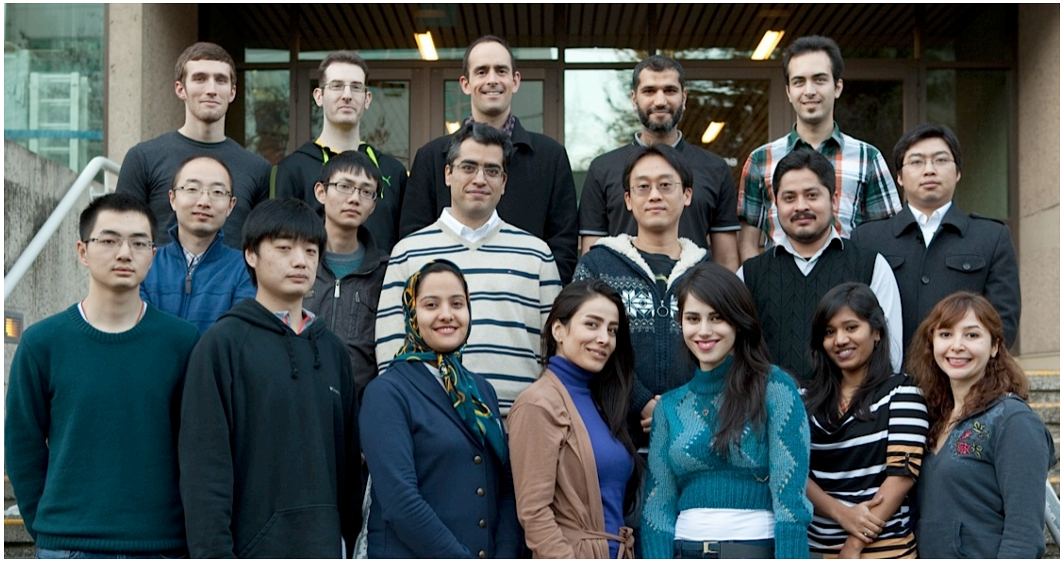




D. Yoo, G. K. Laxminarayana, H.-K. Choi, D. A. Mohr, C. T. Ertsgaard, R. Gordon, S.-H. Oh, "Low-Power Optical Trapping of Nanoparticles and Proteins with Resonant Coaxial Nanoaperture Using 10 nm Gap," Nano Letters 18 (6), 3637-3642 (2018).



bottom-up
natural self-assembly
echnological self-assembly
top-down
UV lithography
electron-beam lithography
na o-imprint lithography
scanning probe lithograp y
proteins, antibodies
viruses
animal cells
small molecules
ribosomes
genes
bacteria
human hair
atoms
DNA ba es
chromosomes
0.1 nm
1 nm
10 nm
100 nm
1 µm
10 µm
100 µm
1 mm
crystalline lattices
carbon nanotubes
quantum dots
ates of transistors
electromechanical, fluidic, optical, magnetic microsystems
DNA microarrays



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