

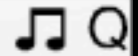
*PhD Scientific Days 2019*

# Manipulating nanoscale systems one by one

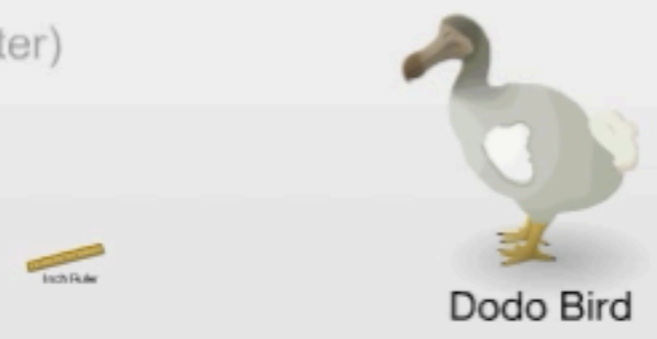
Miklós Kellermayer

Semmelweis University  
Department of Biophysics and Radiation Biology

# Giant Earthworm



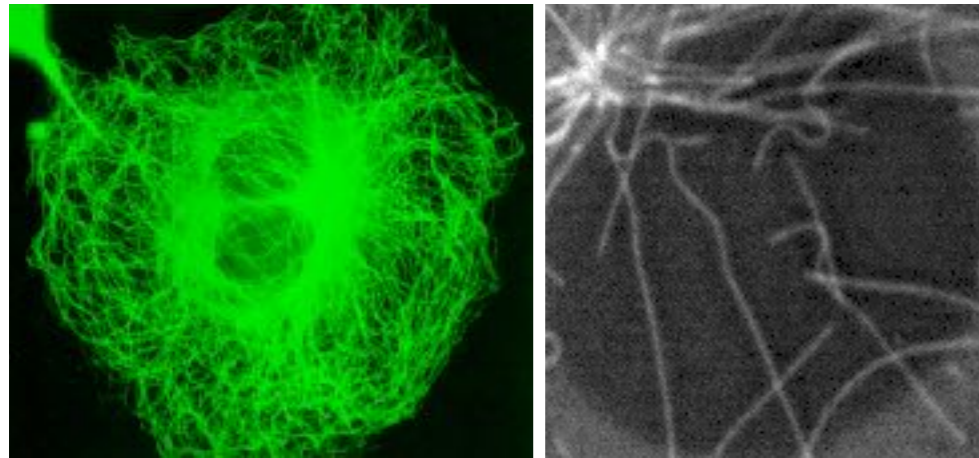
Meter (m) (Diameter)  
 $10^0$  meters



$10^{0.0}$

# Why one by one?

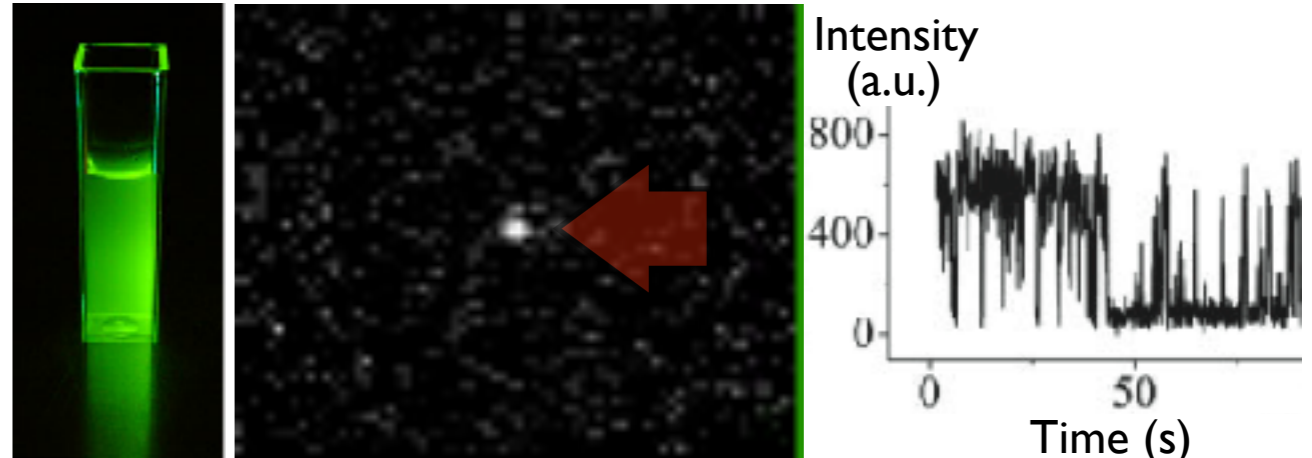
1. Individuals (spatial and temporal trajectories) may be identified in a crowd



Ensemble -  
microtubular system

Single microtubules -  
treadmilling

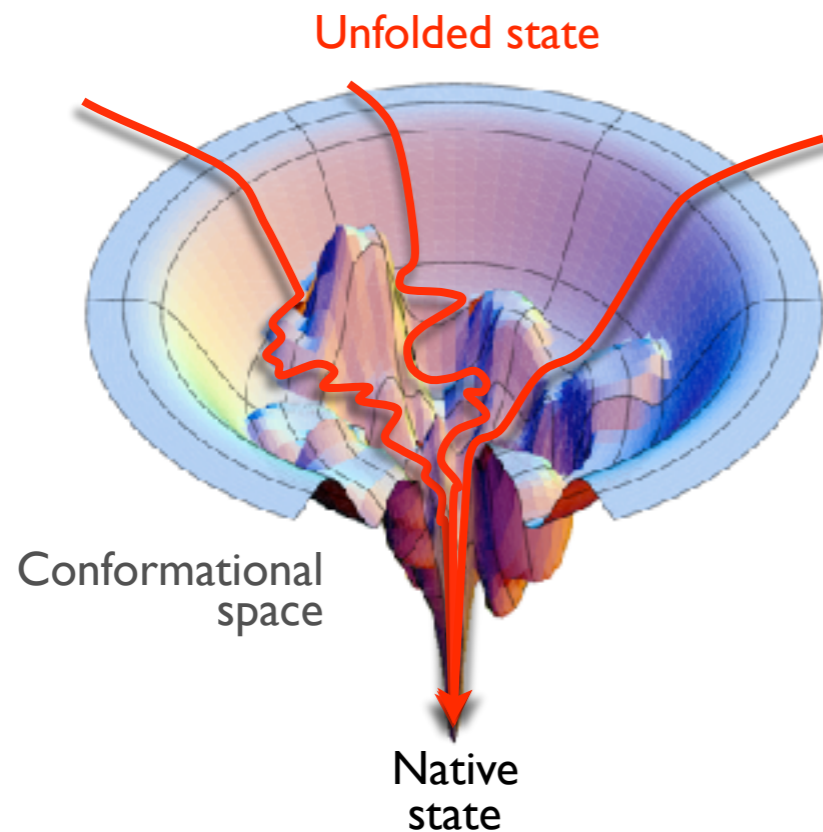
2. Stochastic processes may be uncovered



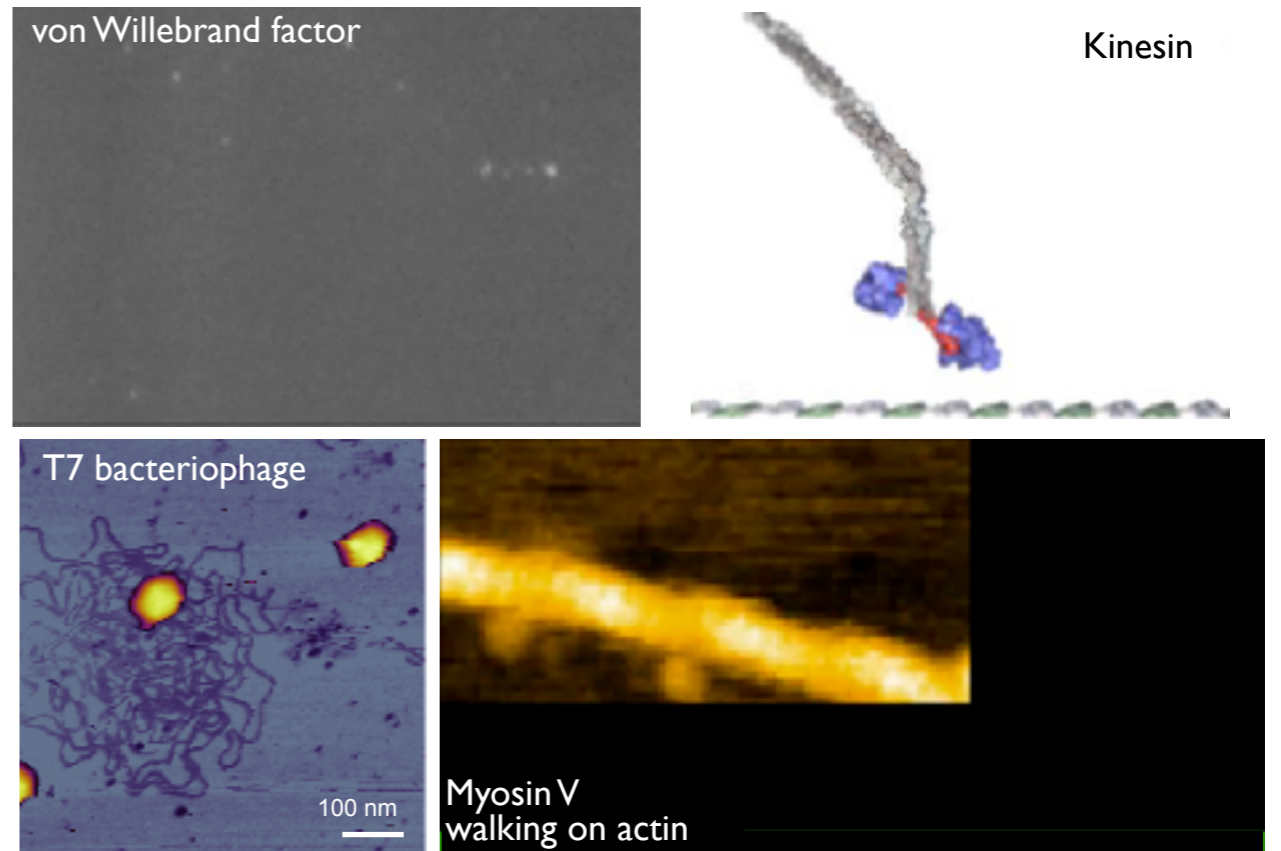
Ensemble -  
intensity

Single quantum dot - blinking

3. Parallel-pathway events may be identified



4. Mechanics of biomolecules may be characterized

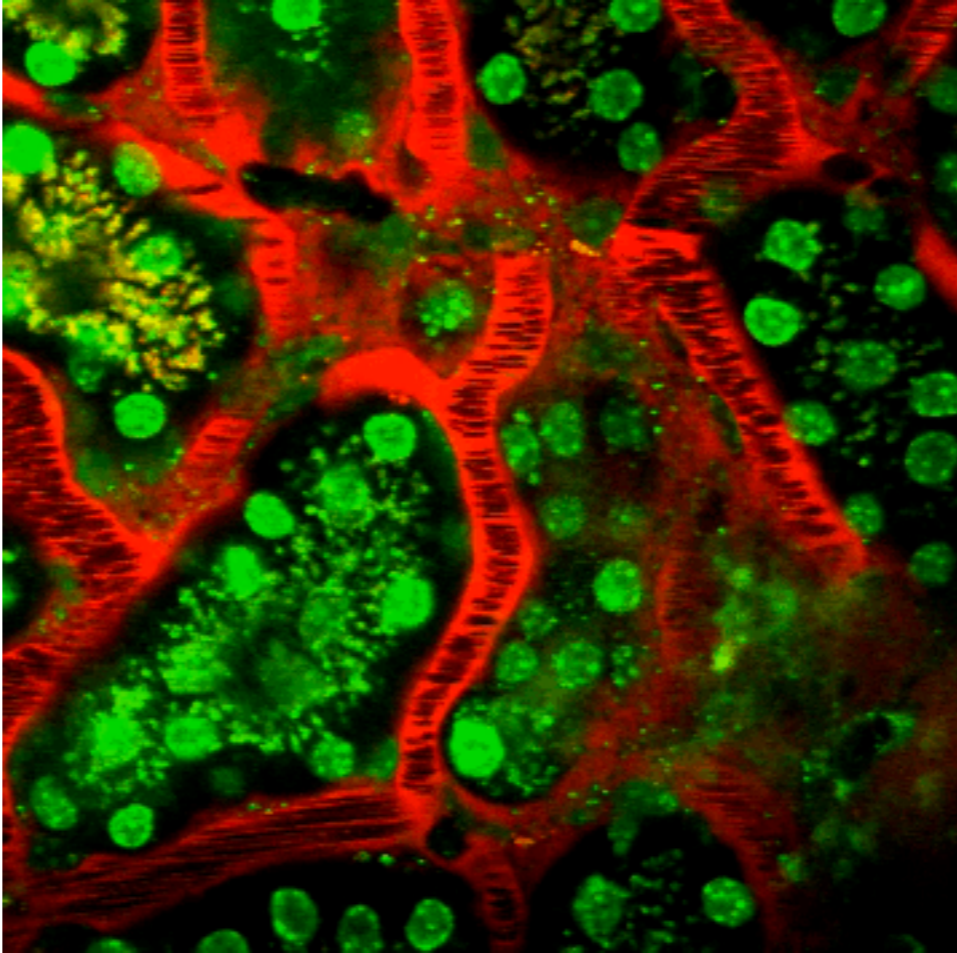
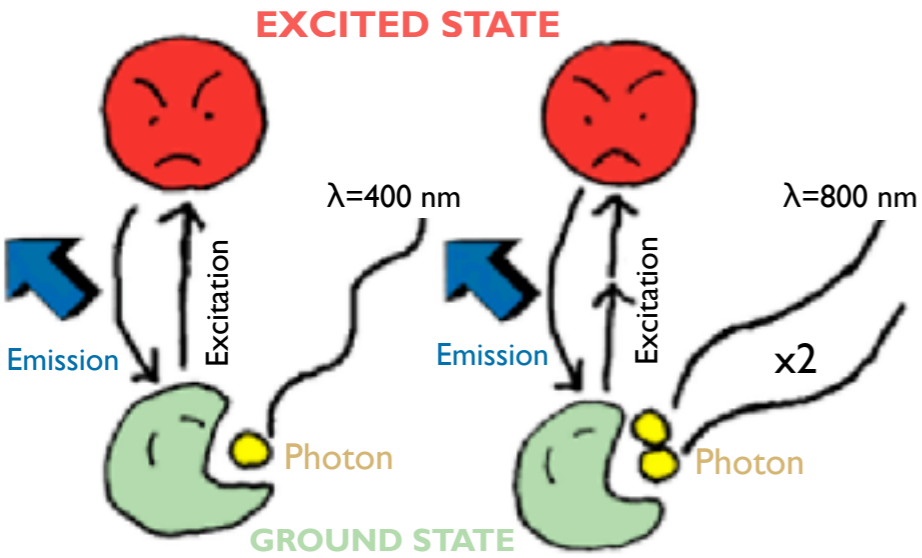




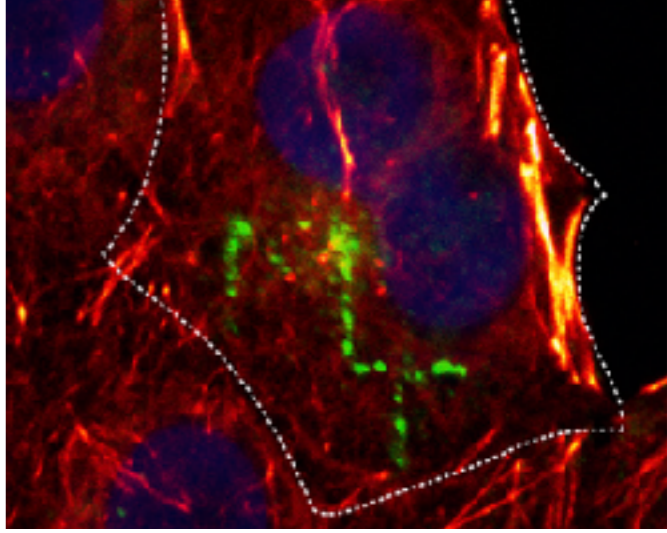
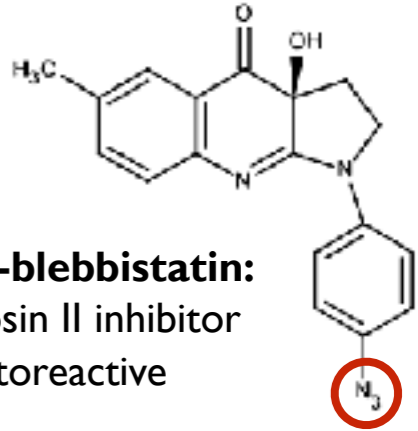
# Manipulation with photochemistry

## Multiphoton microscopy

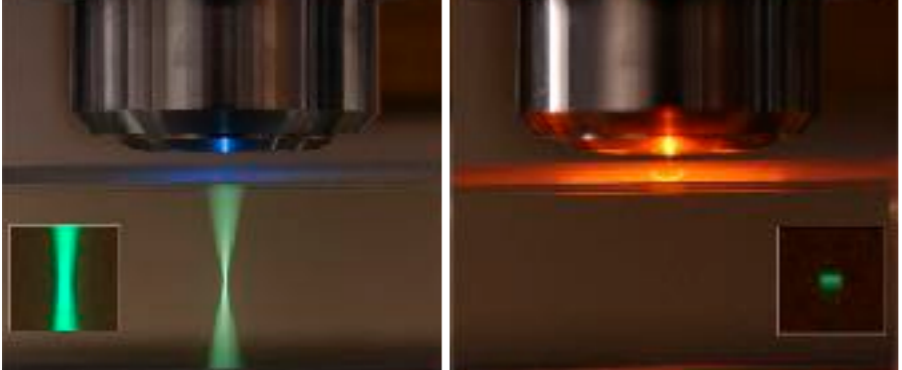
- Photons add up during excitation
- Excitation/emission in focal point
- Excitation with near-IR, fs light pulses
- Large penetration due to long wavelength
- Light-sensitive reactions in fl volumes



Green: kidney tubules; red: albumin



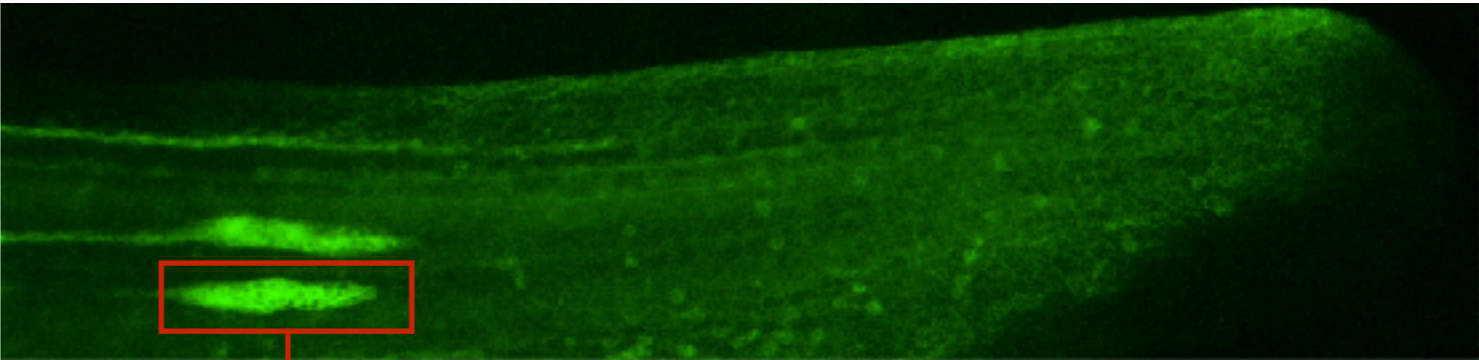
Molecular tattoo with azido-blebbistatin



Single-photon fluorescence

Two-photon fluorescence

Zebrafish lateral line organ development stops upon 2P exposure

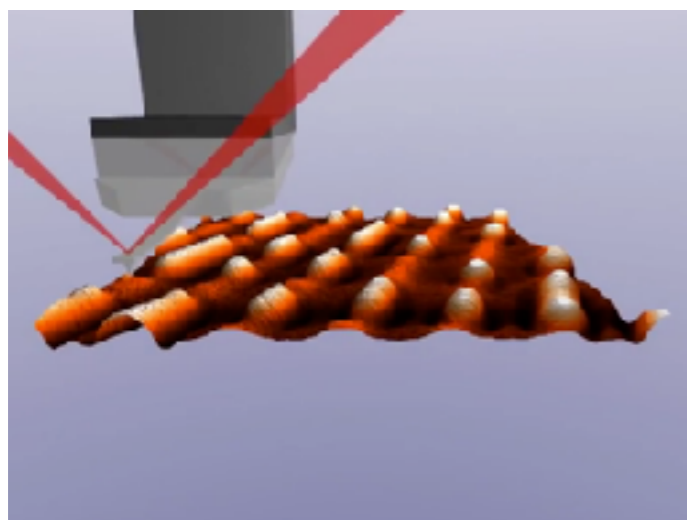
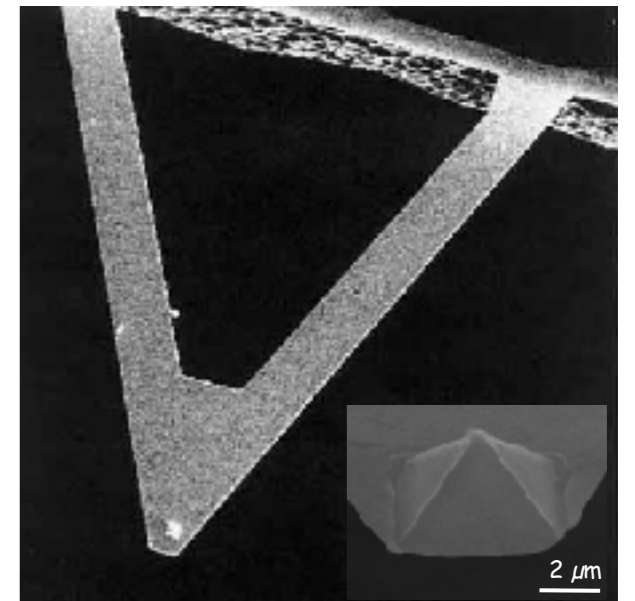
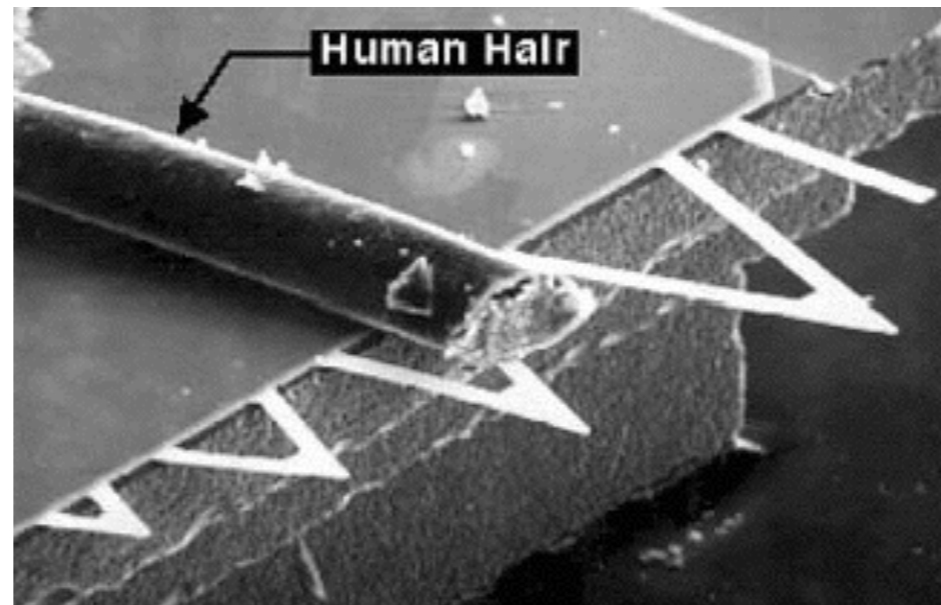
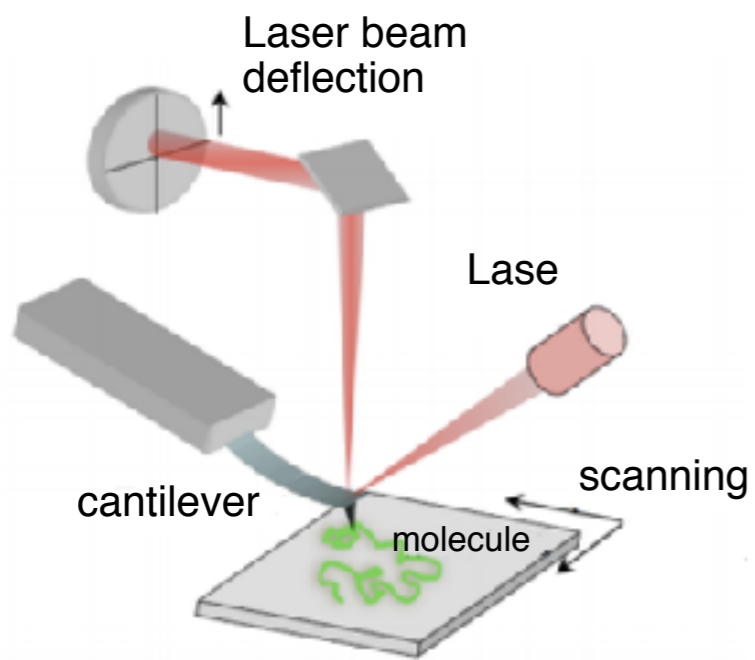


2P exposure

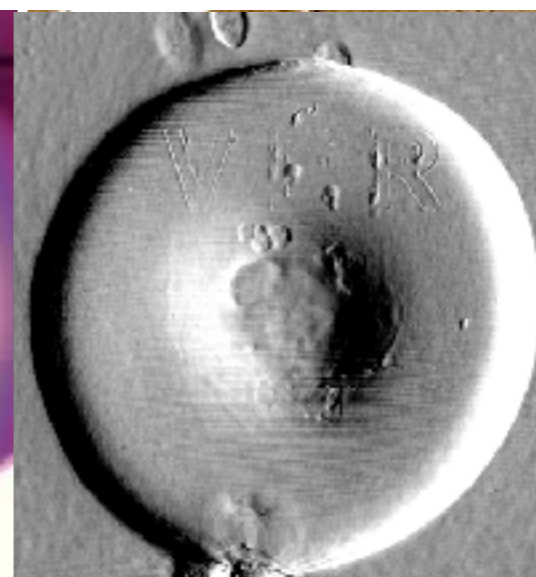


# Manipulation with AFM

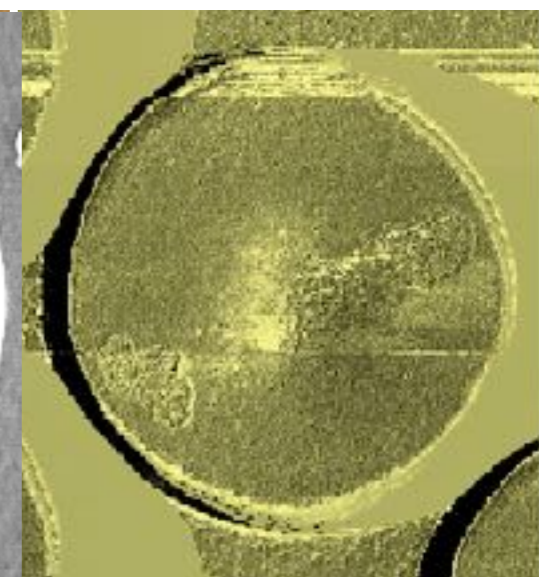
## Atomic Force Microscopy



Height contrast



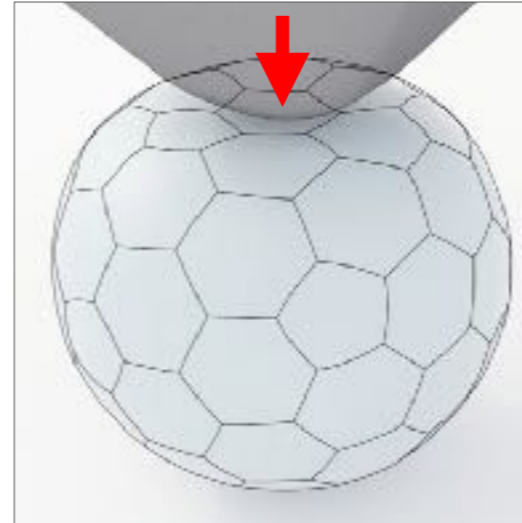
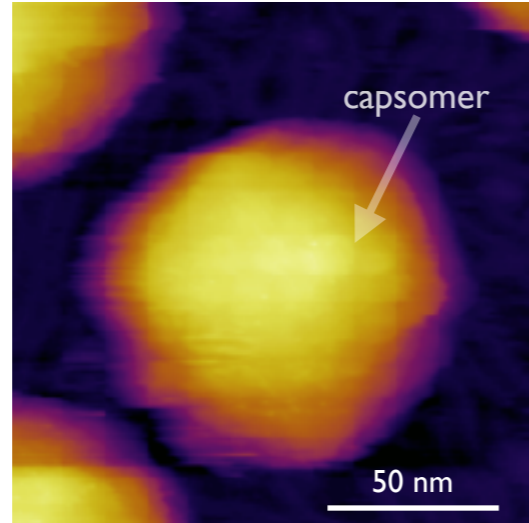
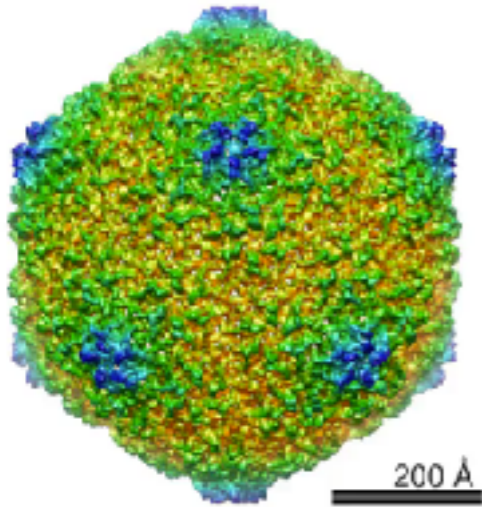
Amplitude contrast



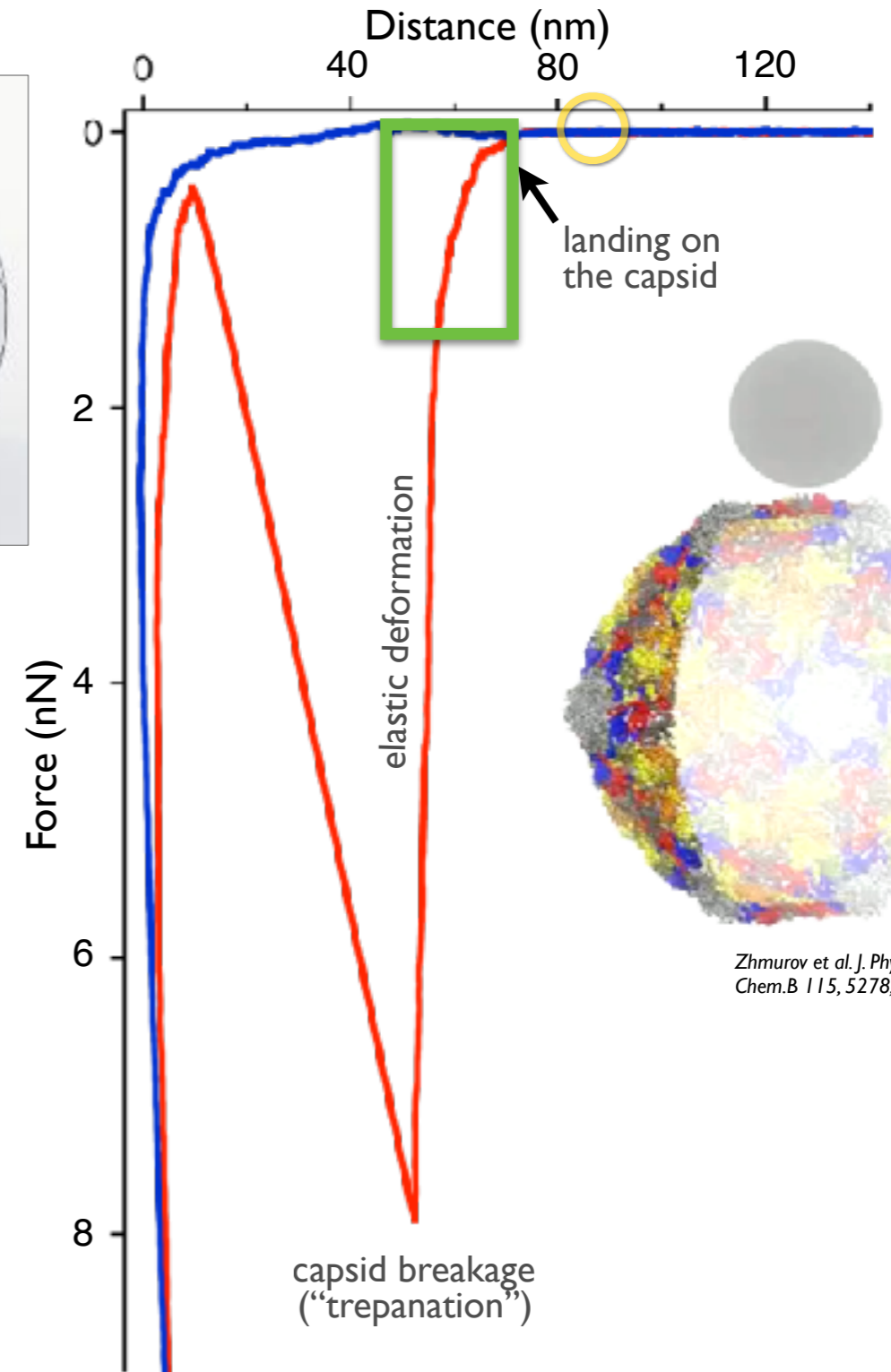
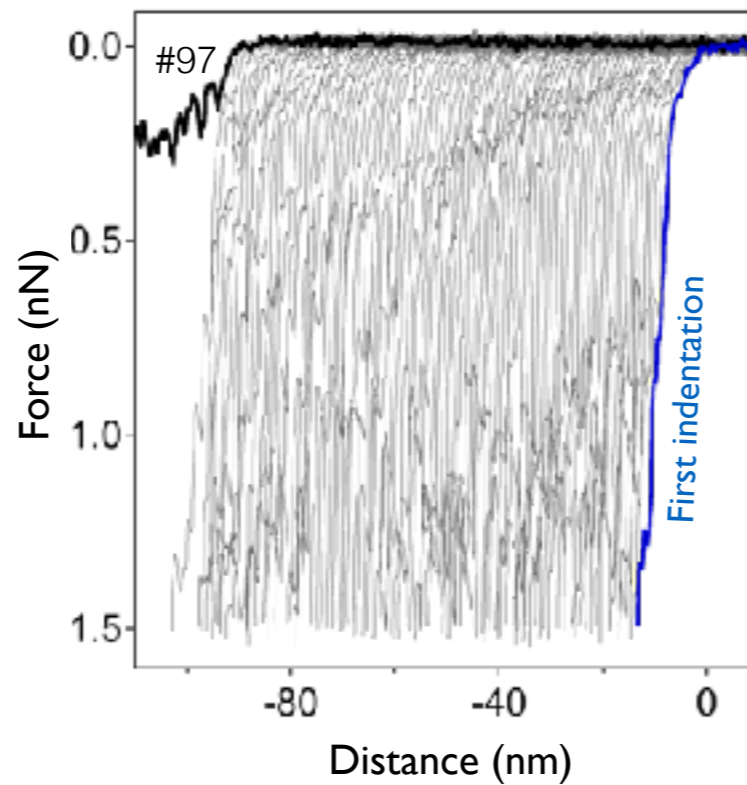
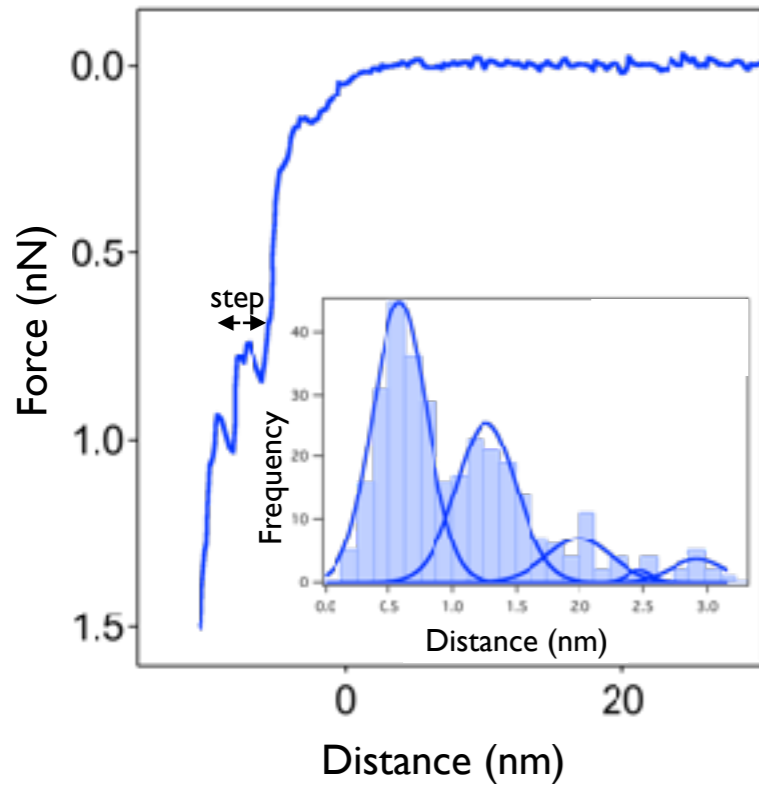
Phase contrast

# T7 phage nanomechanics

Short-tailed icosahedral virus



Ionel et al. *J. Biol. Chem.* 286, 234, 2010.

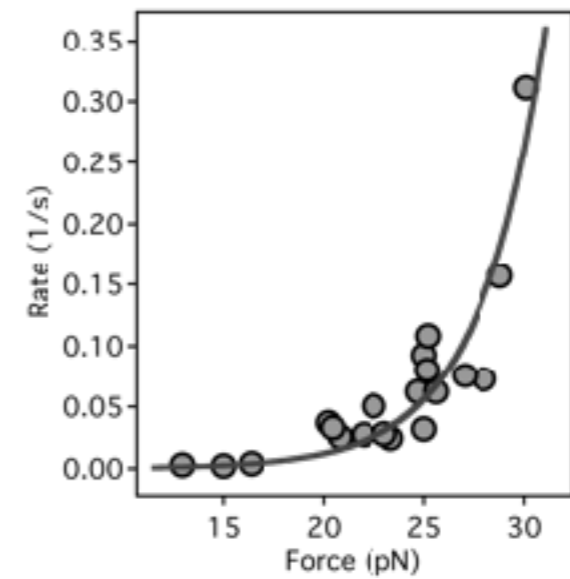
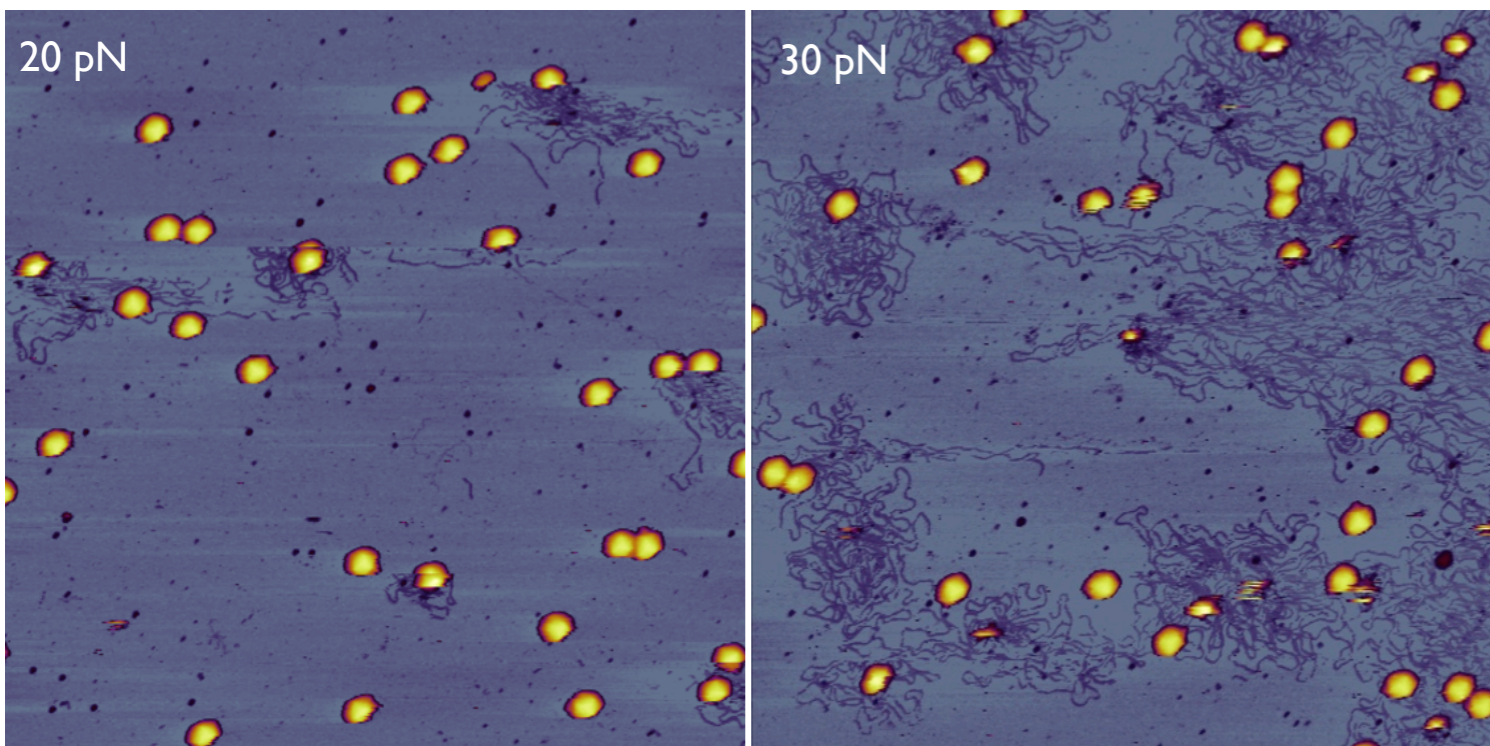
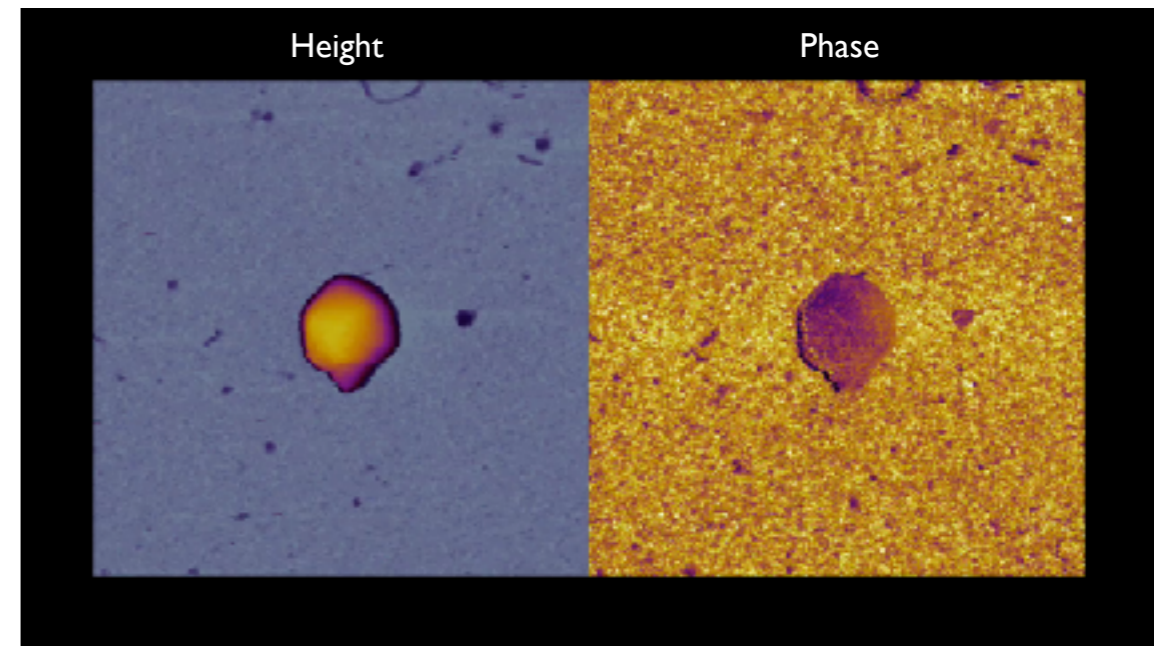
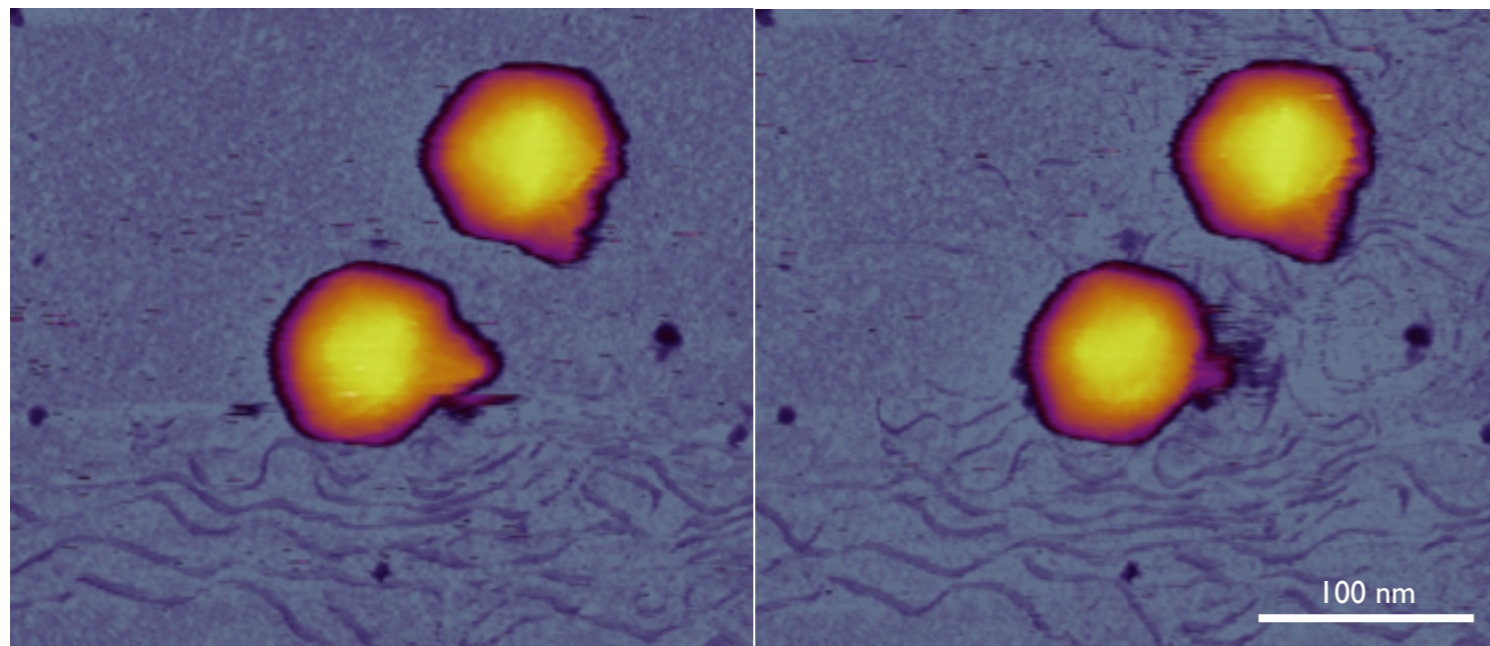


Zhmurov et al. *J. Phys. Chem. B* 115, 5278, 2011.

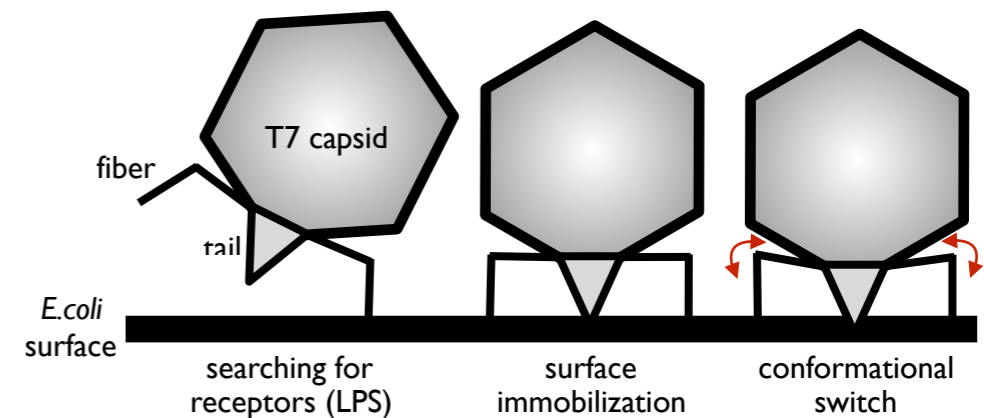
Reversible 6 Å buckling steps, followed by rapid relaxation.



# Mechanically triggered T7 DNA ejection

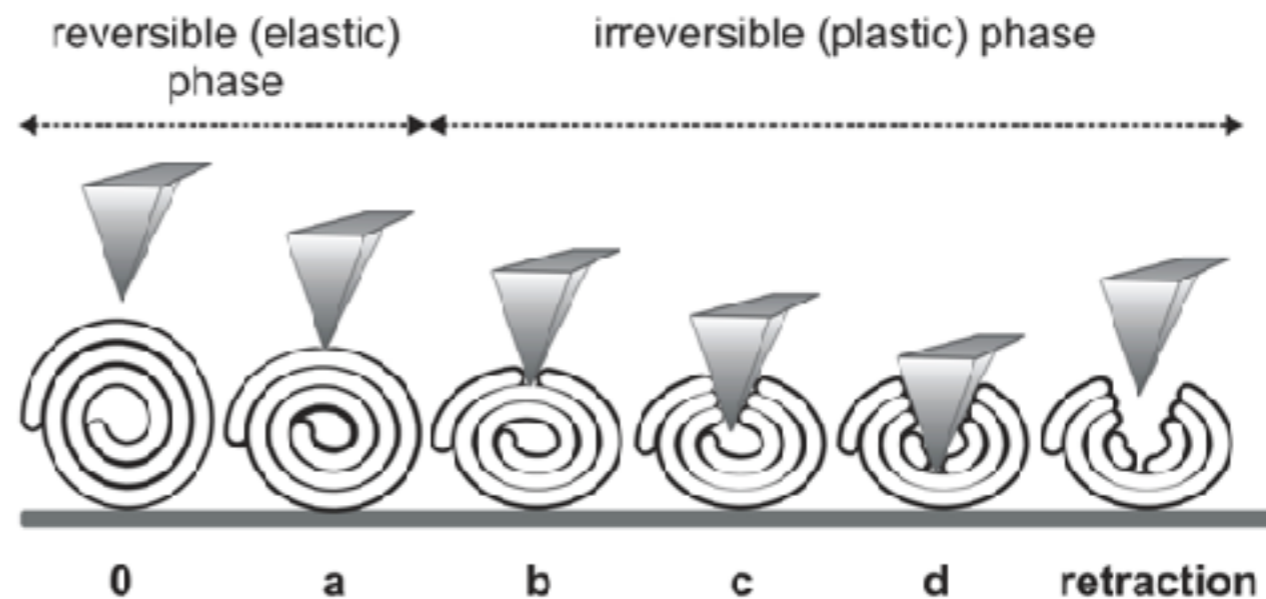
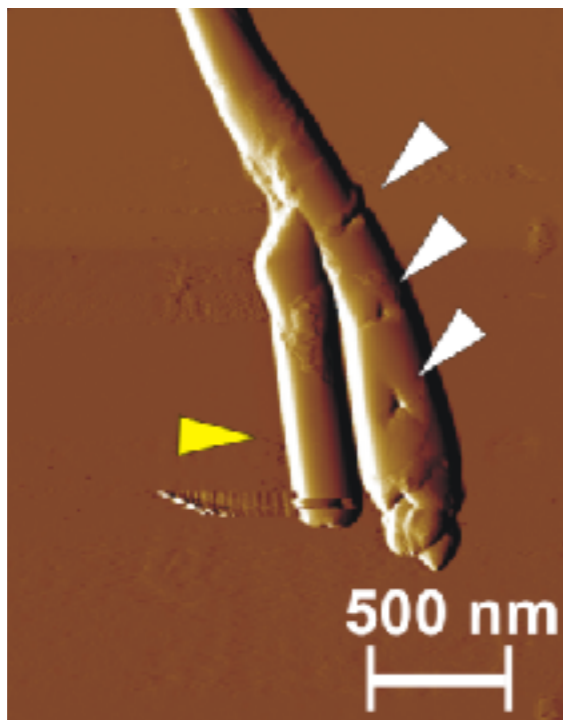
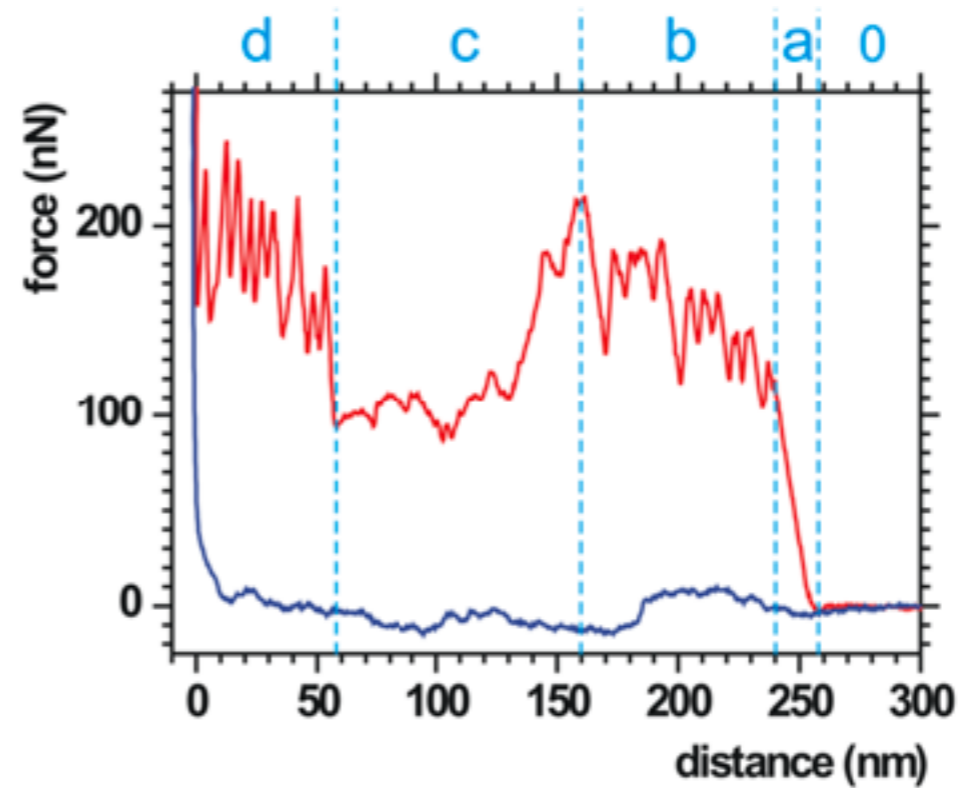
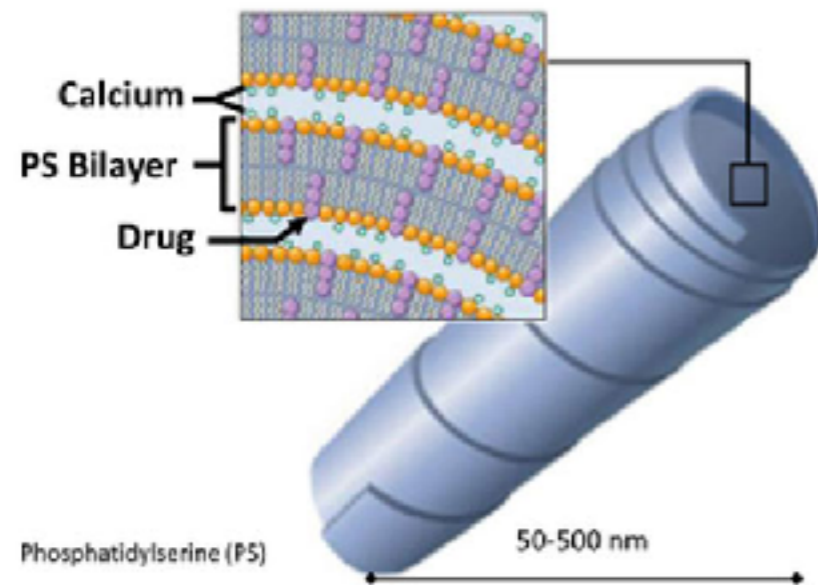


Switch-like trigger response





# Nanomanipulation of cochleates





# Manipulation with light



Optical tweezers



Arthur Ashkin,  
Nobel-prize 2018

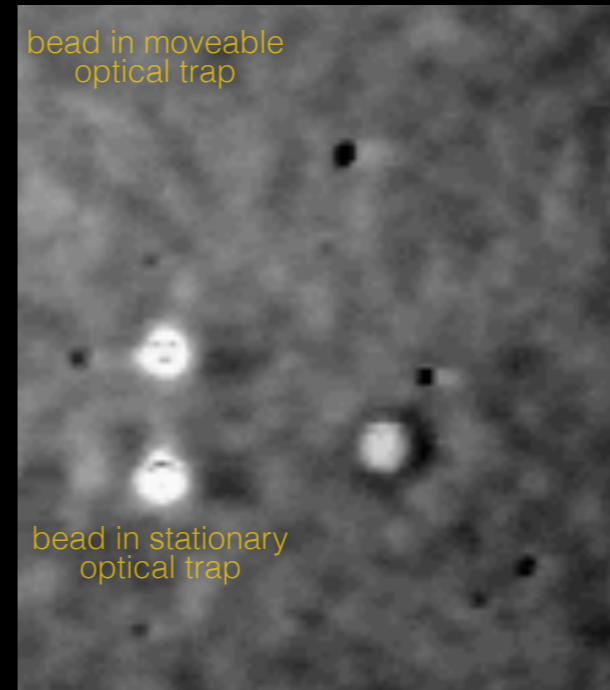
*E. coli* bacterium

Actin filament

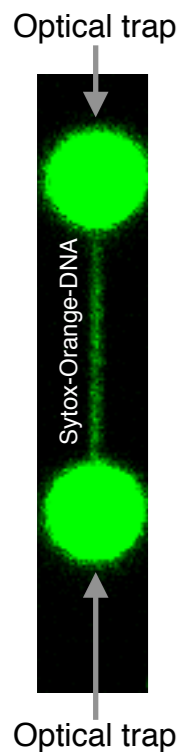
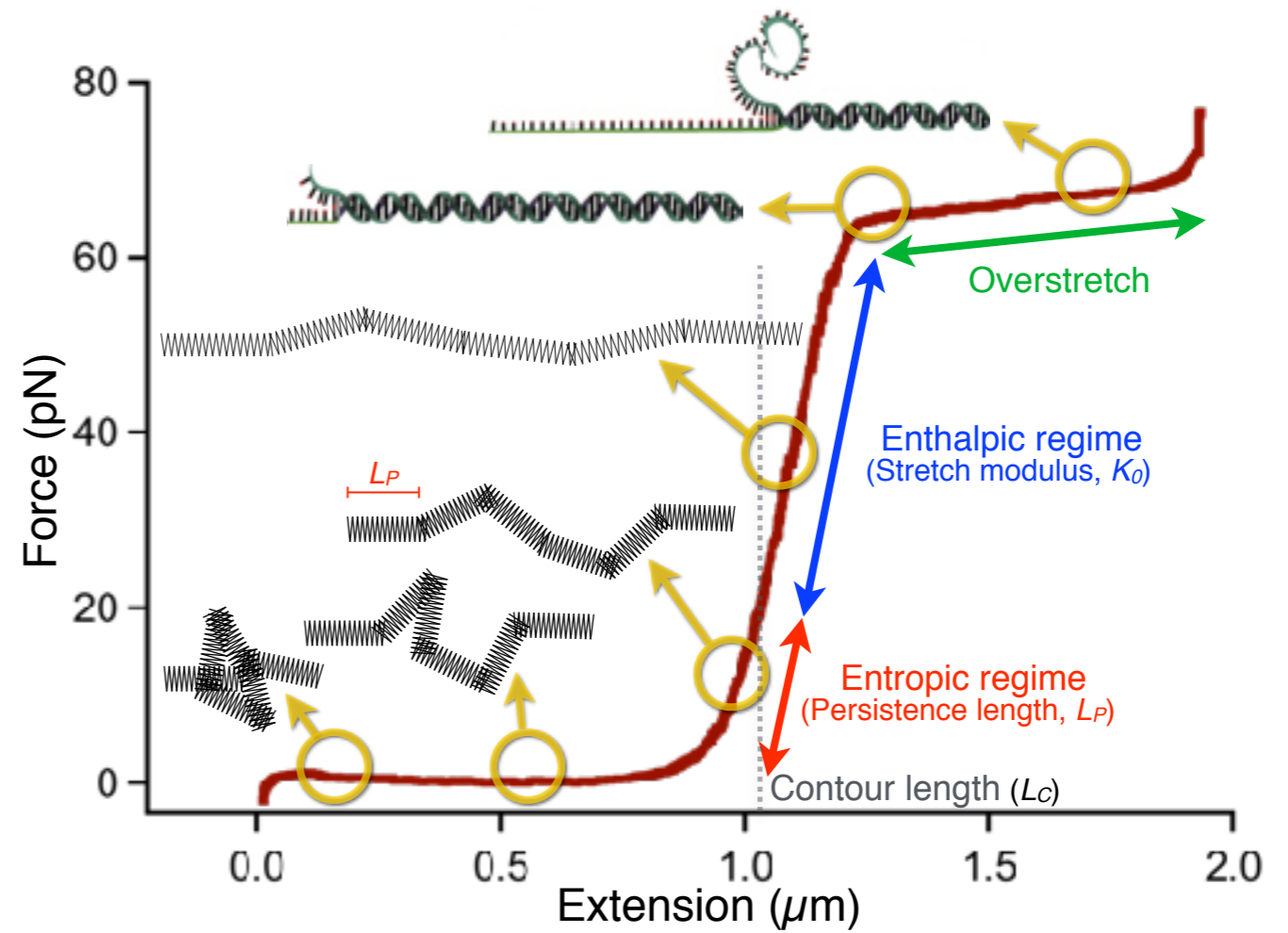
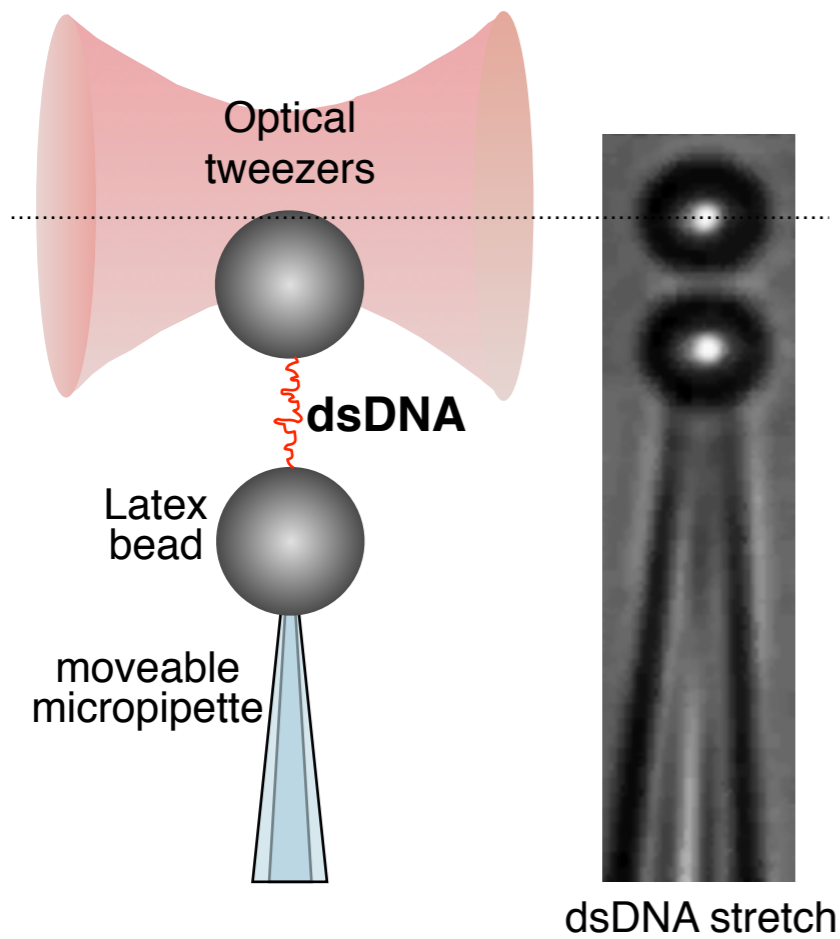
DNA

Phase contrast image

Fluorescence image

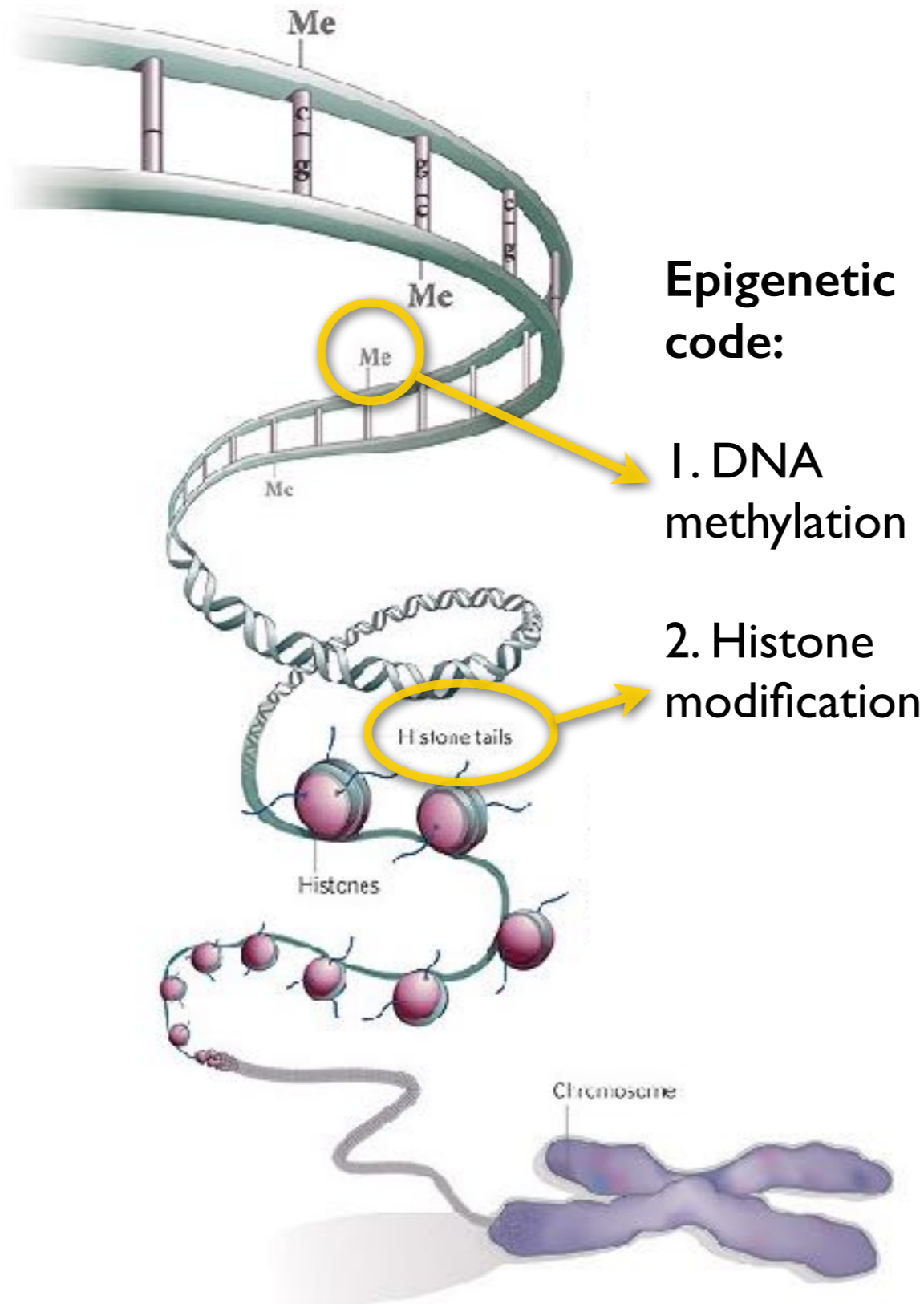


# Mechanical extension of dsDNA





# Do epigenetic modifications alter DNA mechanics?



## Functions of DNA methylation:

### 1. Transcriptional gene silencing

Prader-Willi syndrome

Deletion on paternal chr15



Angelman syndrome

Deletion on maternal chr15



Juan Carreño de Miranda: *The Nude Monster* (1680)

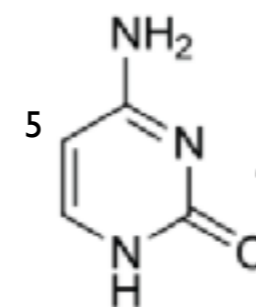
Giovanni Francesco Caroto: *Boy with a Puppet* (1555)

### 2. Genomic stability and protection

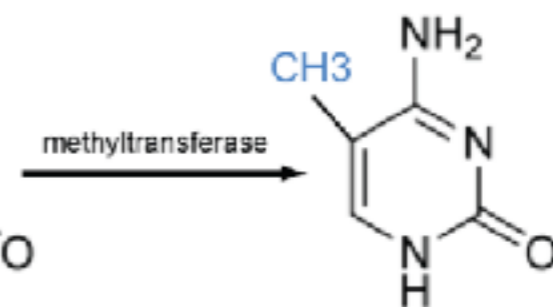
### 3. Chromatin compaction

### 4. Suppression of homologous recombination

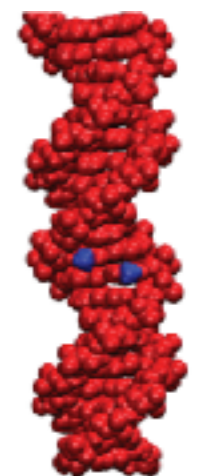
### 5. X-chromosome inactivation (in women)



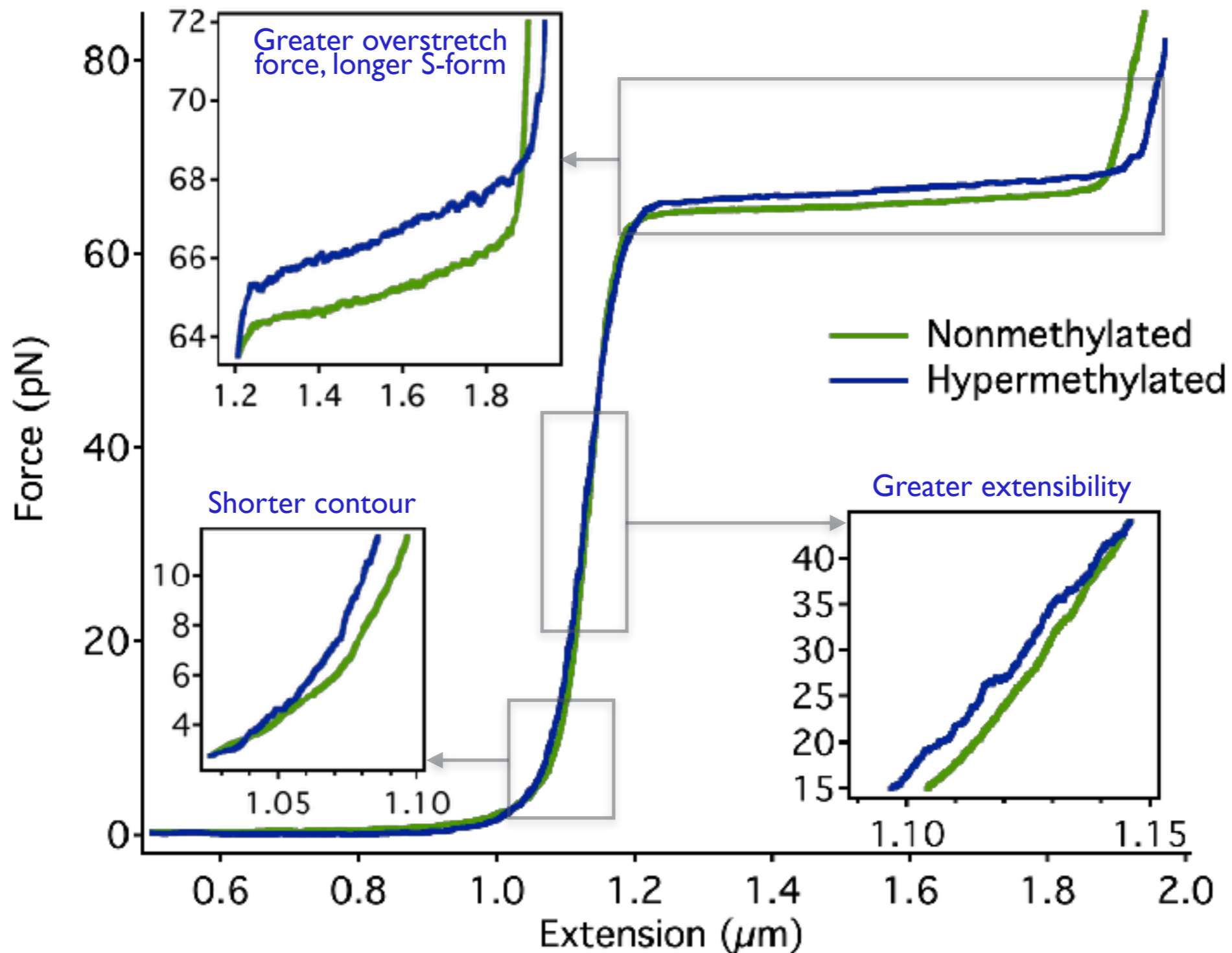
Cytosine in CpG islands



Methyl groups in the large groove of DNA



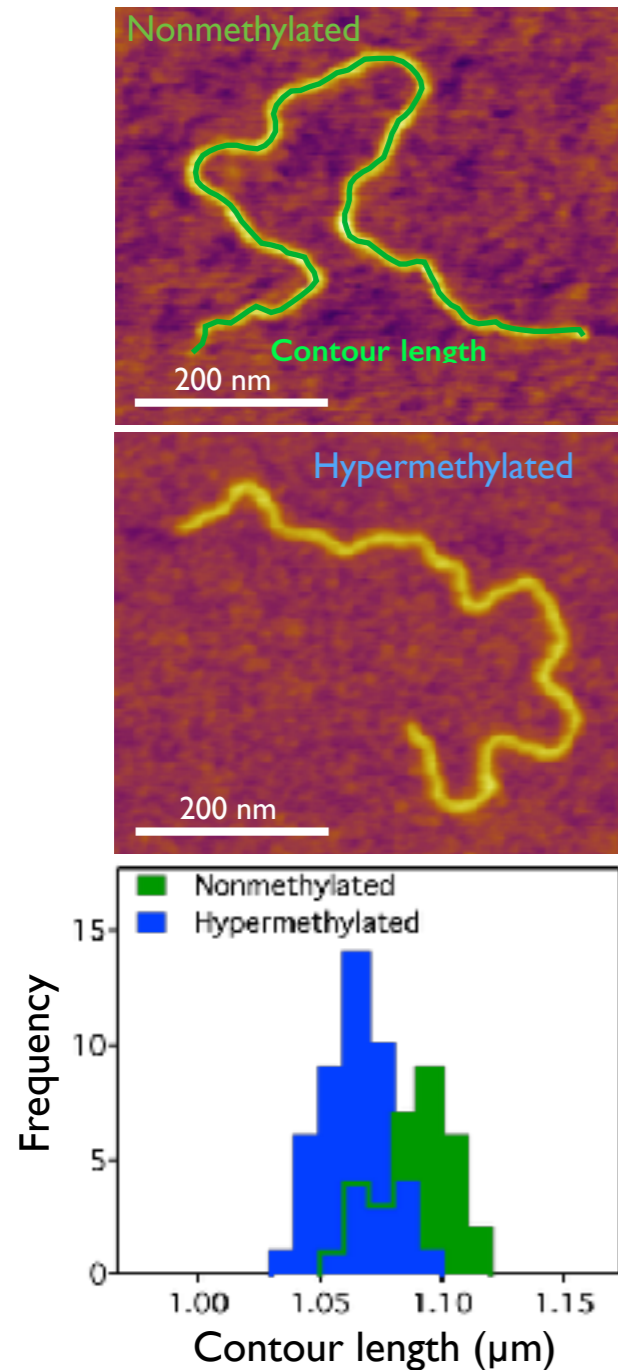
# Nanomechanics of hypermethylated DNA explored with optical tweezers



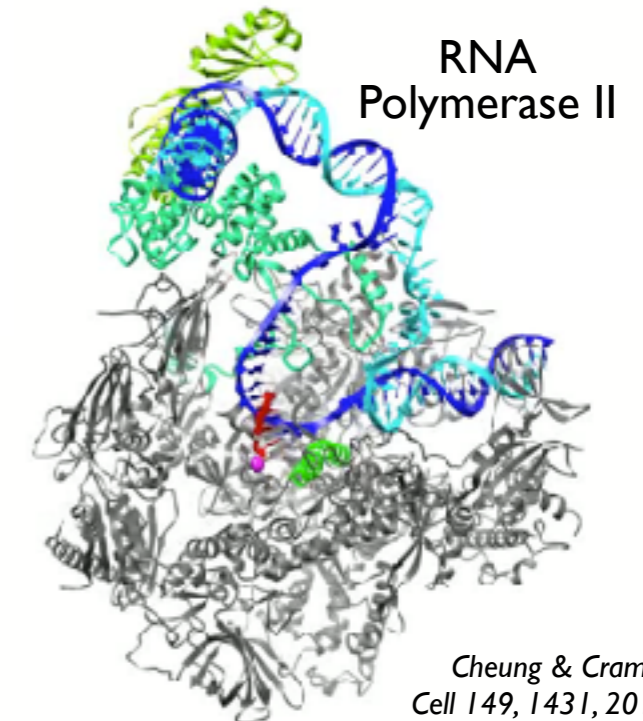
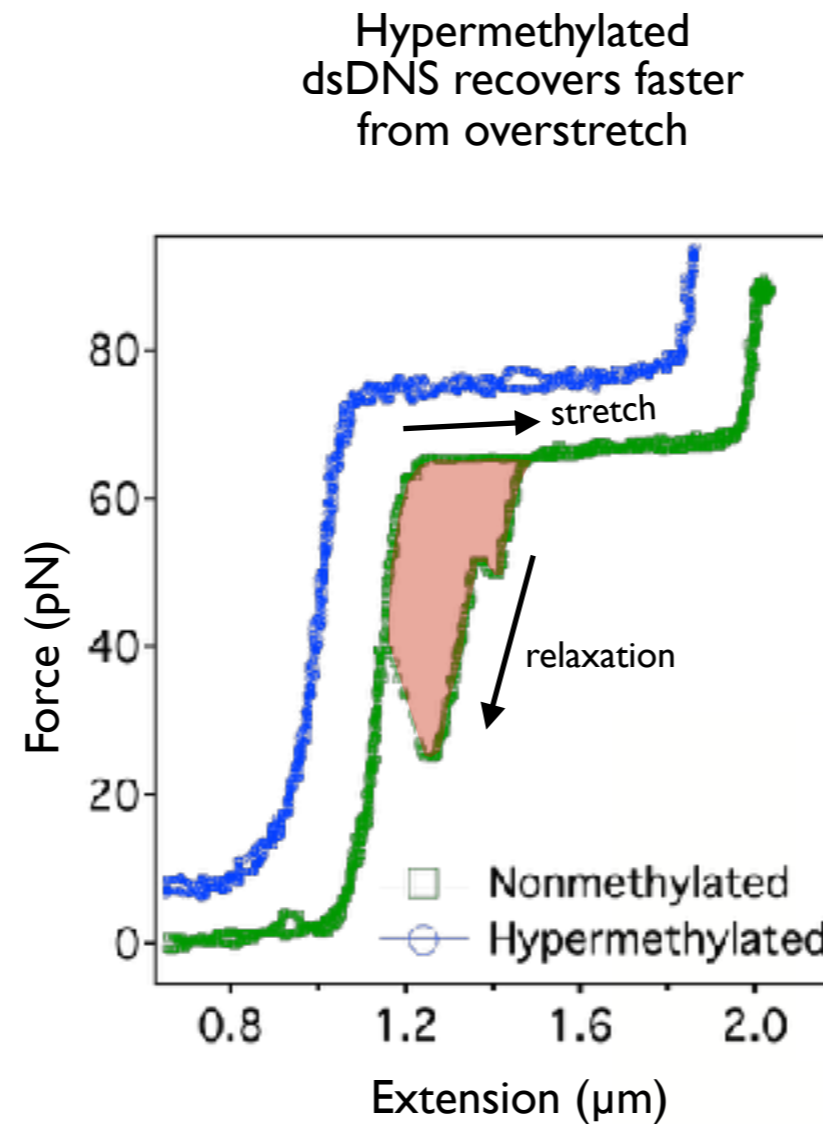


# Hypermethylated dsDNA is more compact, and structurally and mechanically more stable

Structural contour length of hypermethylated dsDNA is shorter

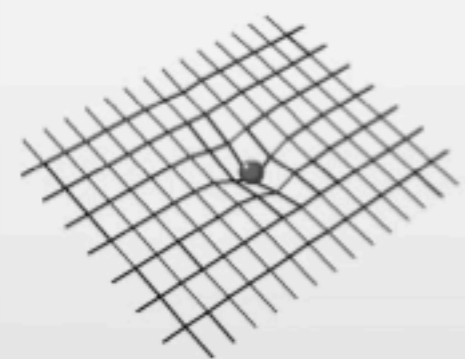
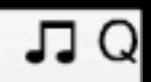


Helicity is retained in hypermethylated dsDNA



Force hysteresis, present in nonmethylated dsDNA, disappears in the hypermethylated form

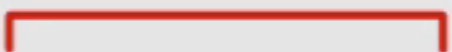
Cheung & Cramer  
*Cell* 149, 1431, 2012



Quantum Foam



String

Planck Length  


$10^{-35.0}$



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Gerald H. Pollack  
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Pályázati támogatás:  
HHMI, MTA, OTKA  
FP5, FP7, ETT, TÉT  
TÁMOP, GVOP, KFKT,  
KTIA, VKSZ, VEKOP,  
NVKP

