ICTS-NCBS-MBI program

Mechanical Manipulations and Responses at the Scale of Cells and beyond

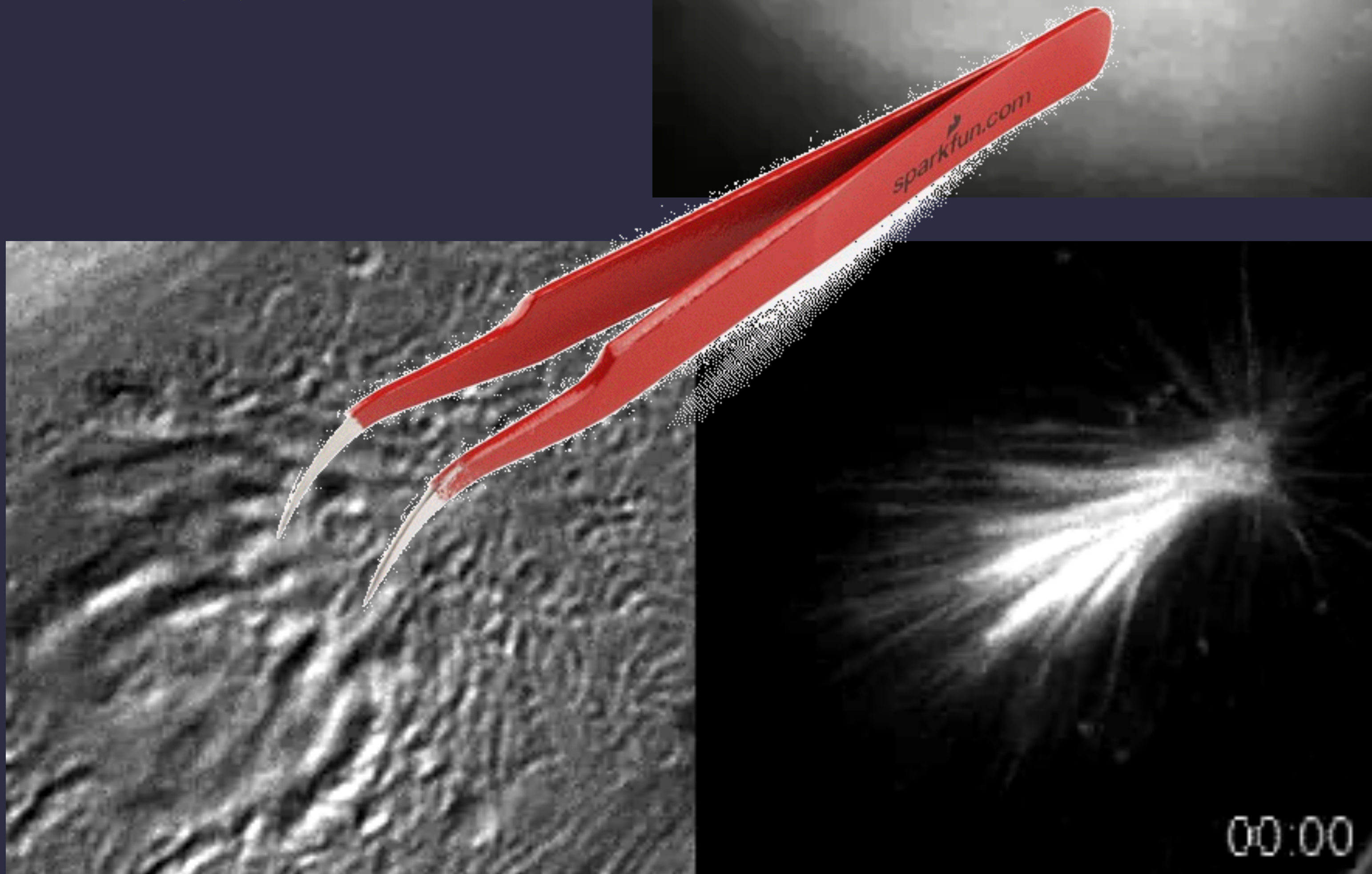
Optical Tweezers

Christoph Schmidt

Georg August University Göttingen

mitotic spindle in
Xenopus oocyte extract,

tubulin speckle
fluorescence imaging (Tarun Kapoor)



In his textbook “A treatise on electricity and Magnetism” (§ 793) (1873) Maxwell tells us:

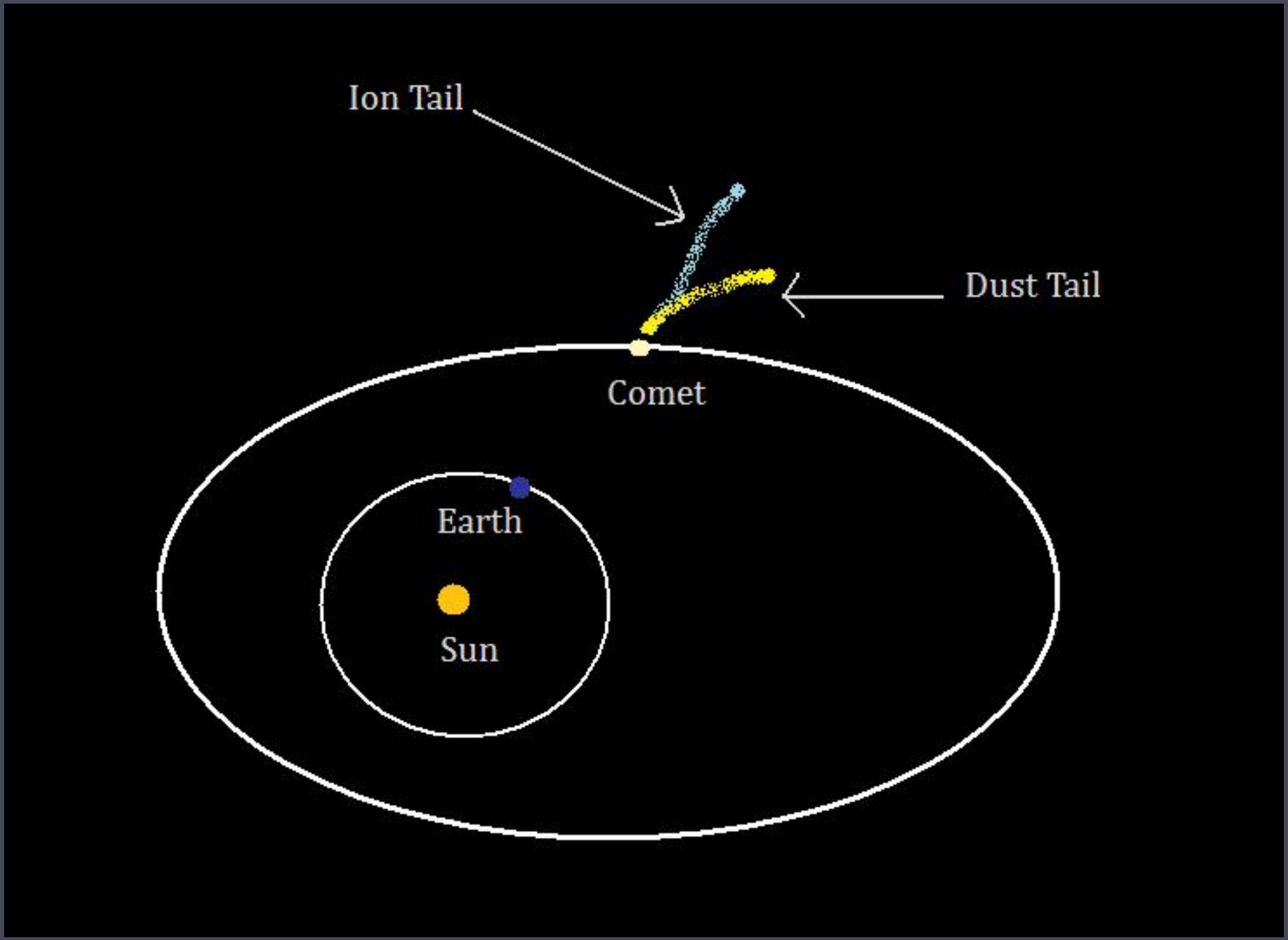
It is probable that a much greater energy of radiation might be obtained by means of the concentrated rays of the electric lamp (than solar light).

Such rays falling on a thin metallic disk, delicately suspended in a vacuum, might perhaps produce an observable mechanical effect.

Estimate: force on a $1 m^2$ plate in full sun light:

0.4 mg if absorbing, 0.8 mg if reflecting

Light pressure on comet tails



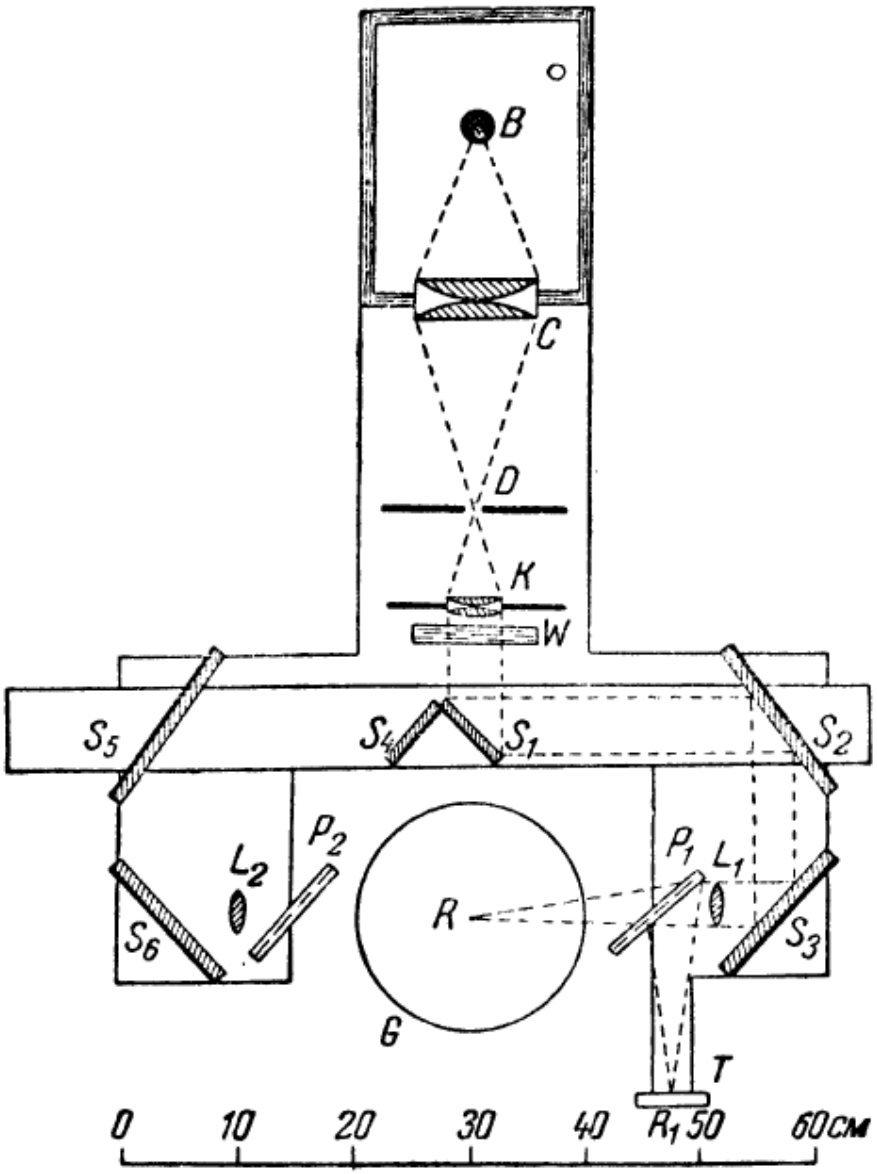
Light pressure on comet tails, Hale-Bopp, 1997

Crookes' radiometer or light mill
Sir William Crookes, 1873

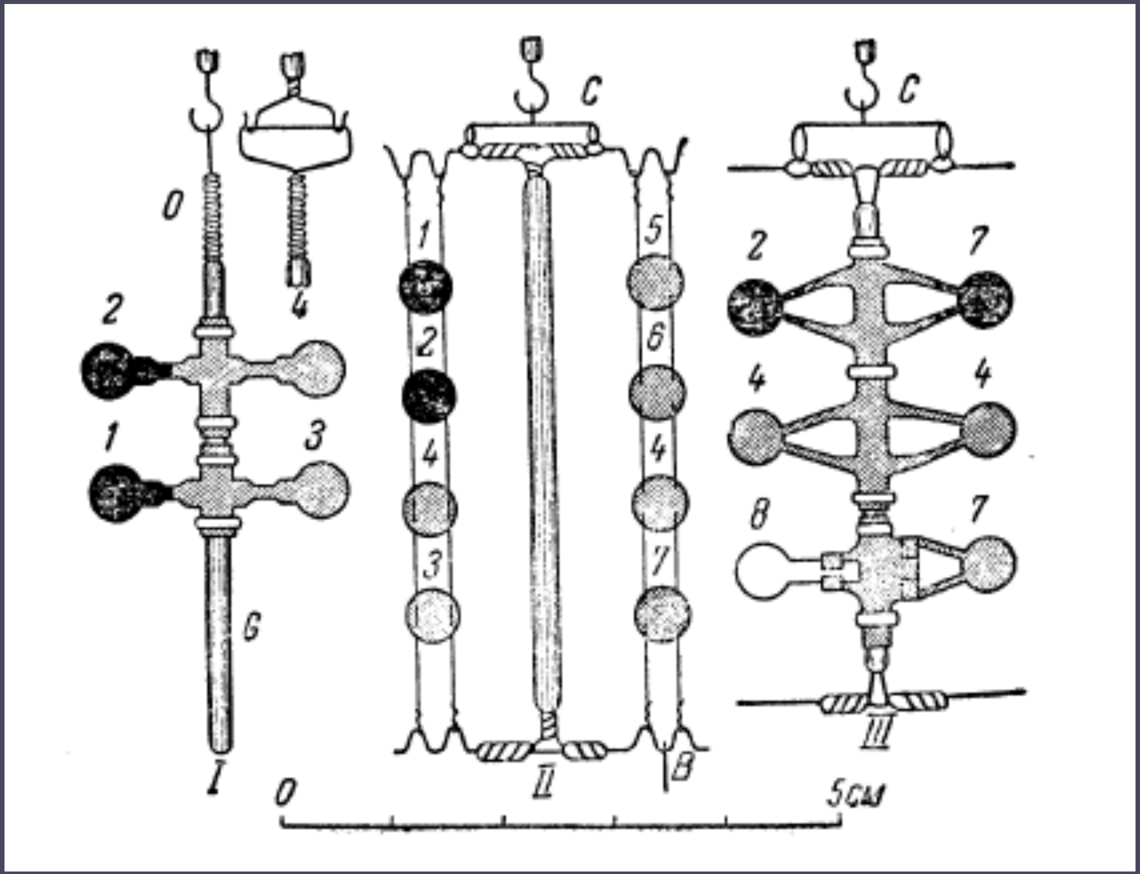
Light pressure experiment 1901:

Pyotr Nikolaevitch Lebedev, Moscow State University

1866-1912



Lebedev’s light pressure experiment: wings used



The results obtained can be stated as follows:

1) The impinging bundle of light yields pressure both on reflecting, and on absorbing surfaces; these ponderomotive forces are not connected with already known secondary convectional and radiometric forces called by heating up.

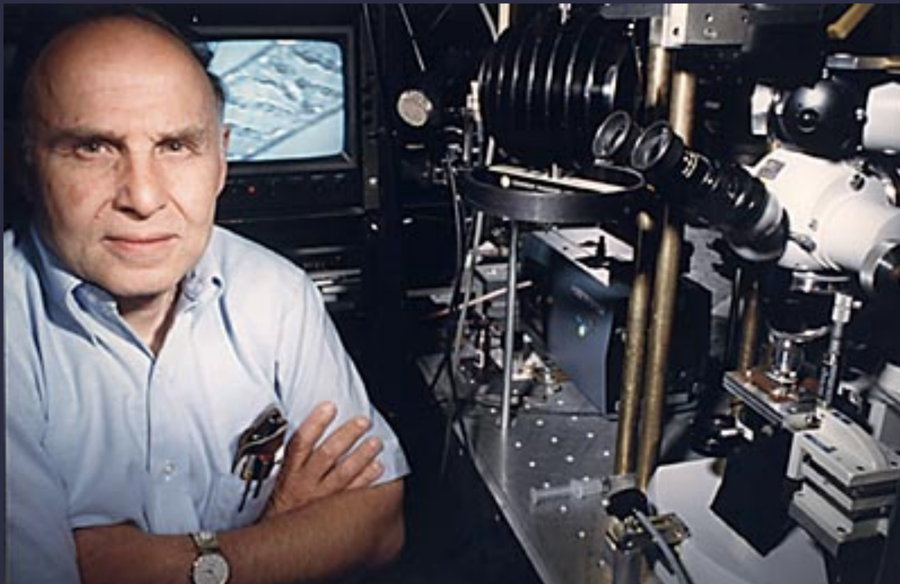
2) The forces of light pressure are directly proportional to the energy of an impinging beam and do not depend on its colour.

3) The observed forces of light pressure, within limits of observational errors, are quantitatively equal to the Maxwell-Bartoli forces of pressure of a radiant energy.

Thus the existence of the Maxwell-Bartoli forces of pressure has been established for the light beams experimentally.

Physical laboratory of the University.

Moscow, August 1901.



Arthur Ashkin, Bell Labs

The Nobel Prize in Physics 1997

"for development of methods to cool and trap atoms with laser light"

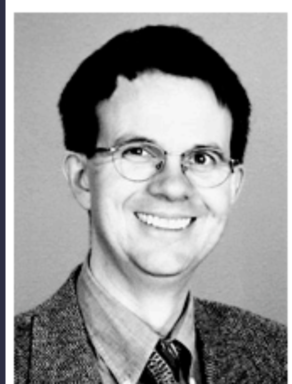
Steven Chu

Claude Cohen-Tannoudji

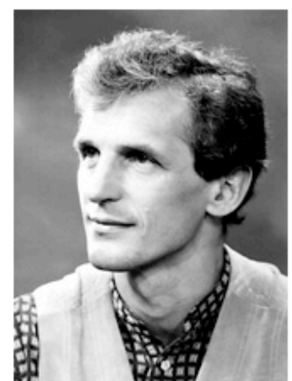
William D. Phillips

The Nobel Prize in Physics 2001

"for the achievement of Bose-Einstein condensation in dilute gases of alkali atoms, and for early fundamental studies of the properties of the condensates"



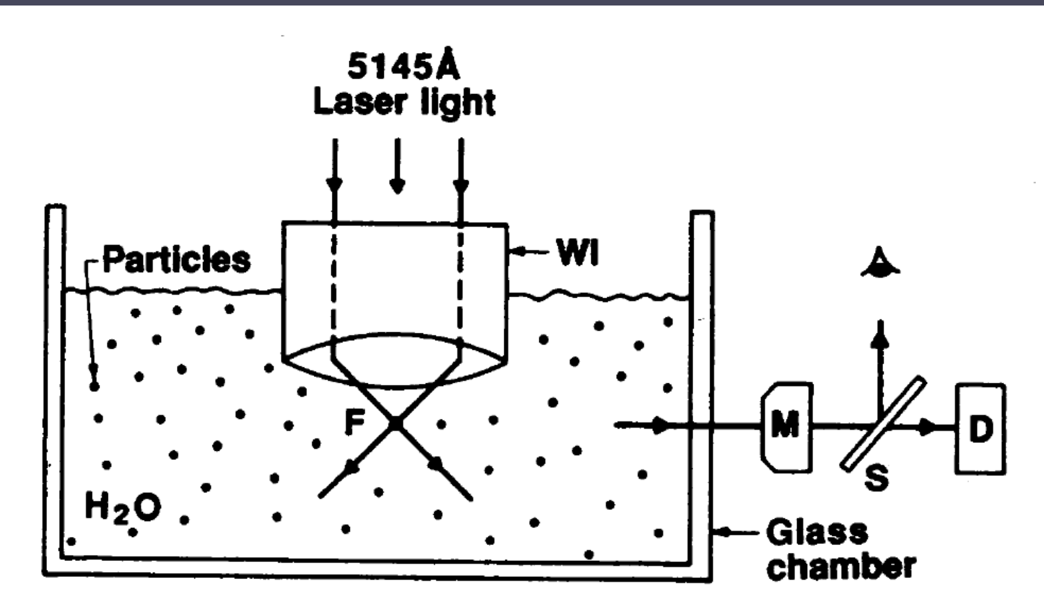
Eric A. Cornell

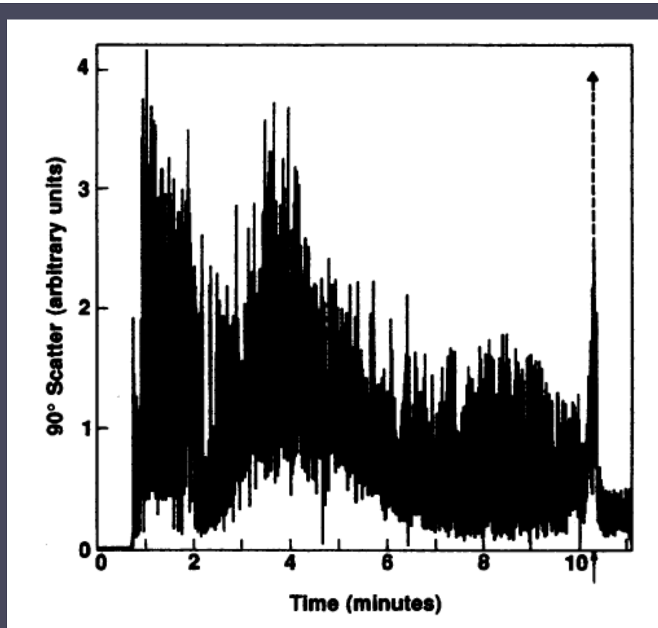


Wolfgang Ketterle

Carl E. Wieman

Ashkin, experiments with TMV virus, Science 1987



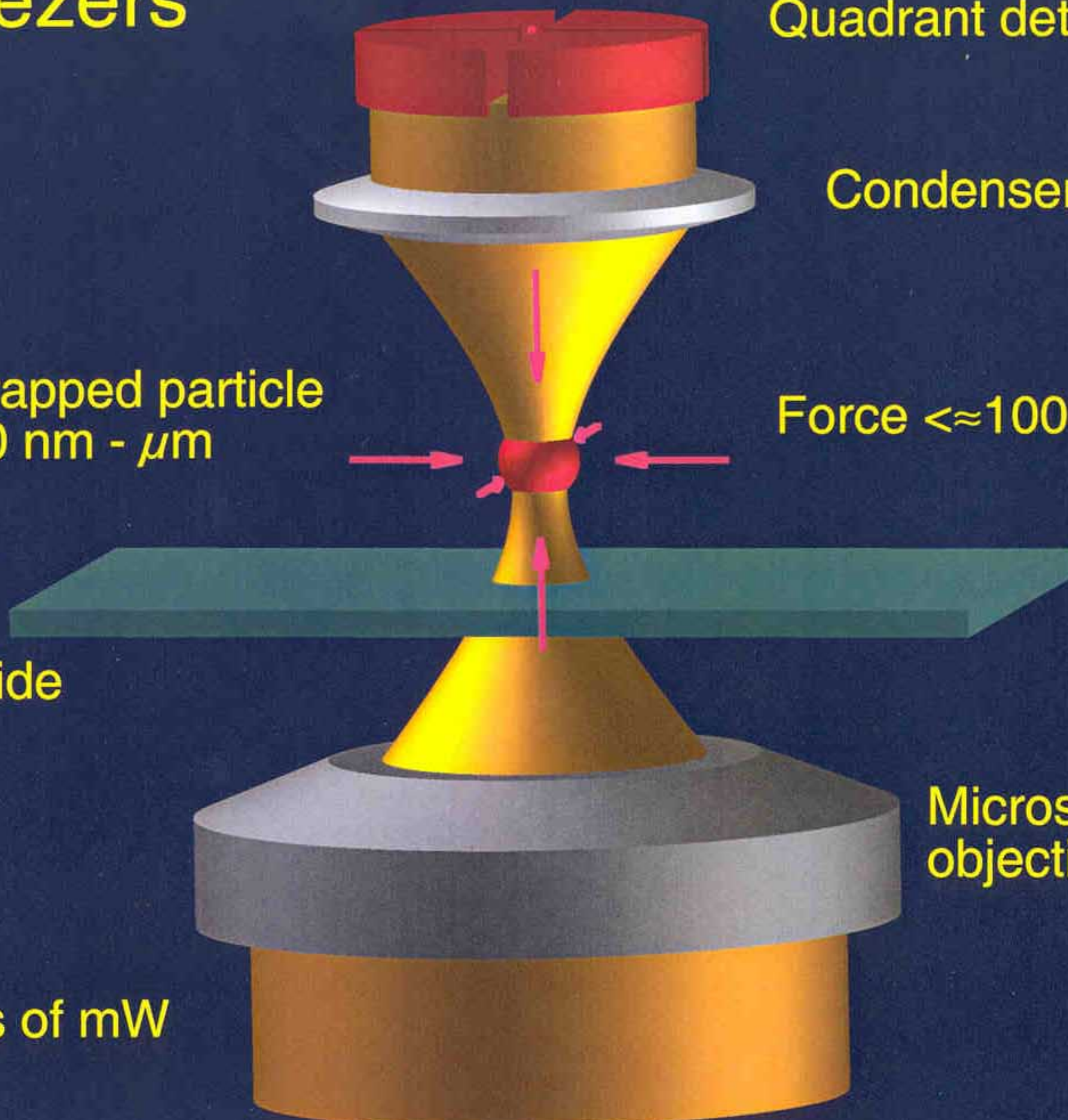


In most of our experiments with silica colloids or TMV in water, we noticed the appearance of some strange new particles in diluted samples that had been kept around for several days. They were quite large compared to Rayleigh particles, on the basis of their scattering of light, and were apparently self-propelled. They were clearly observed moving through the distribution of smaller slowly diffusing Rayleigh-sized colloidal particles at speeds as high as hundreds of micrometers per second. They could stop, start up again, and frequently reversed their direction of motion at the boundaries of the

optical trapping of 0.2 μm silica beads in water

optical trapping of *E.Coli* bacteria in water

Optical Tweezers



Laser beam -> 100s of mW

First suggested: Art Ashkin, 1978, PRL40:729, first realized: Ashkin et al., 1986, Opt. Lett. 11: 288

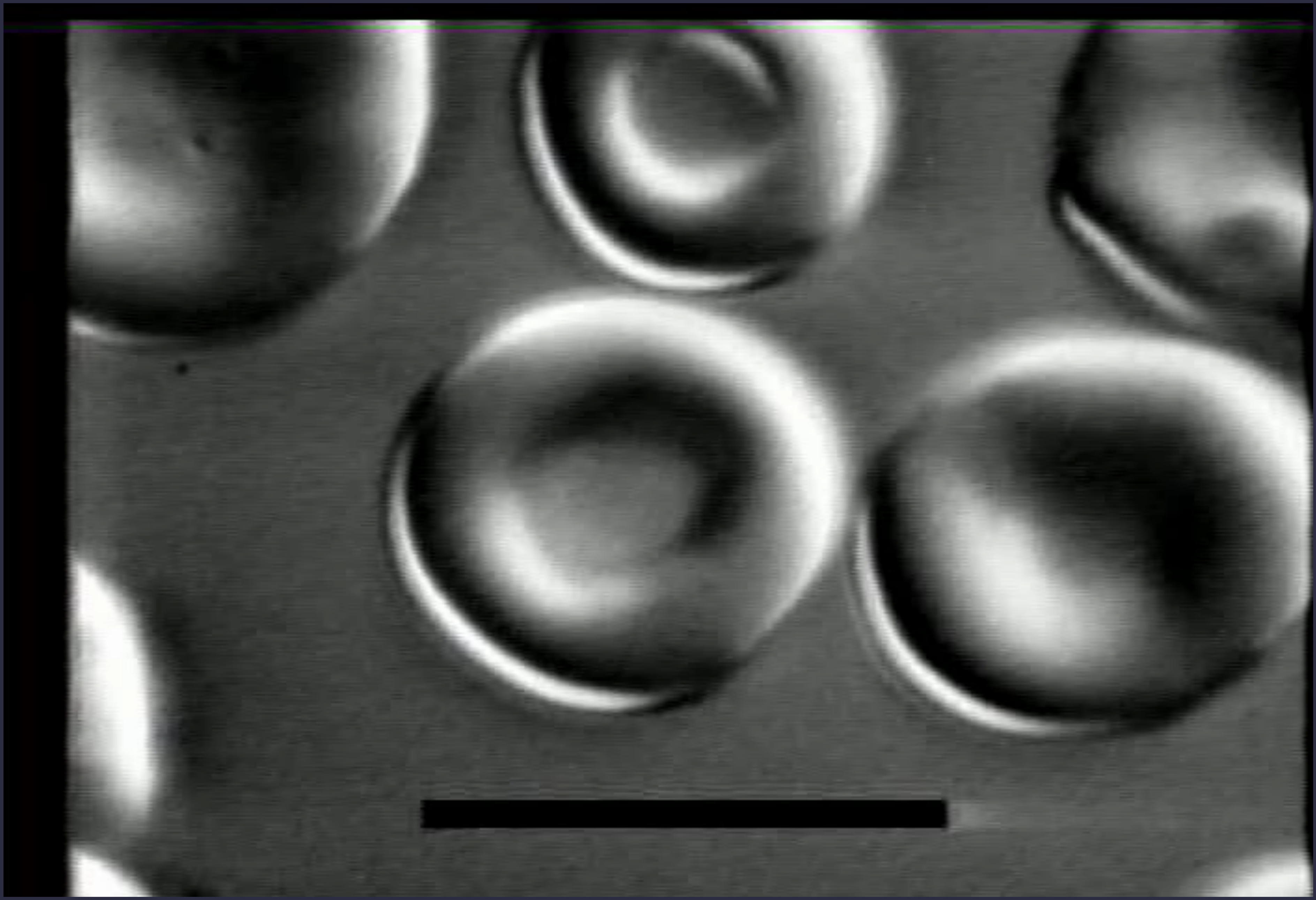
optical trapping

- application of forces up to ~100 pN
- measurement of forces from ~1 pN
- measurement of displacement from ~1 nm

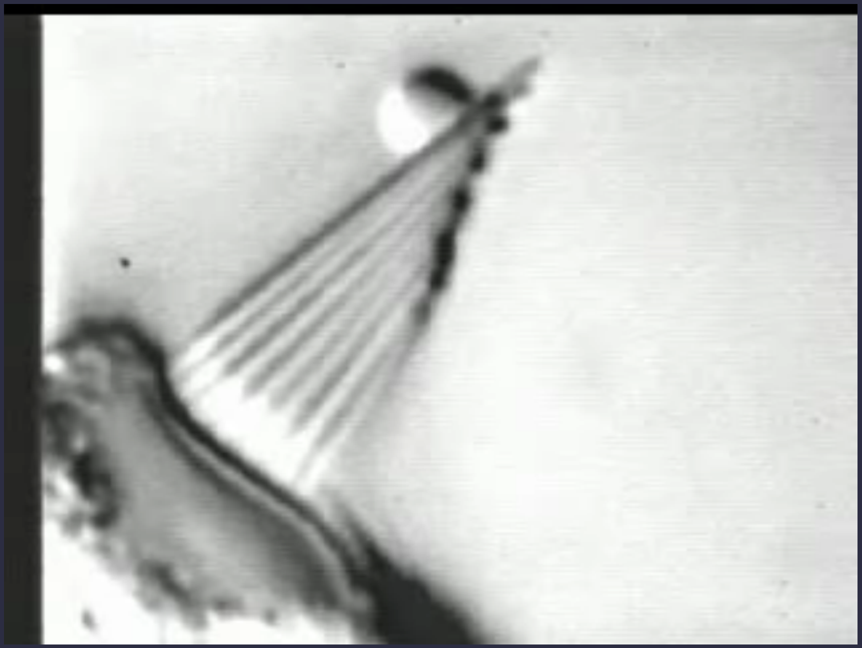
- measurement of force generation / displacement by motor proteins
- manipulation of cells / biomolecules
- measurement of rheological properties of complex fluids

some issues

- thermal (Brownian) noise needs to be dealt with
- optical damage: heating
- trapping causes extra bleaching of fluorescence
- high-frequency detection needs the right detectors

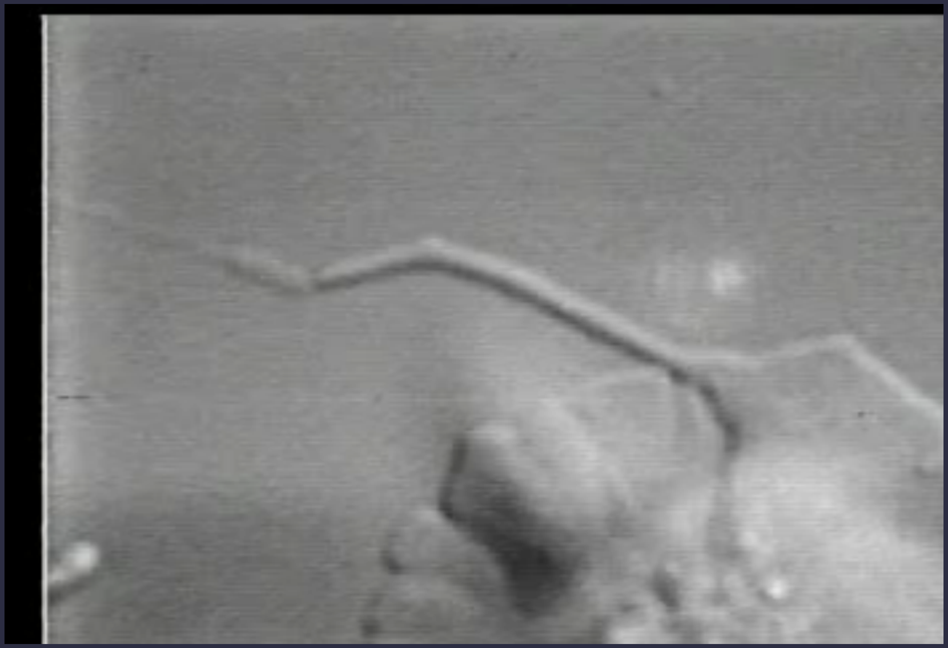


manipulating parts of cells

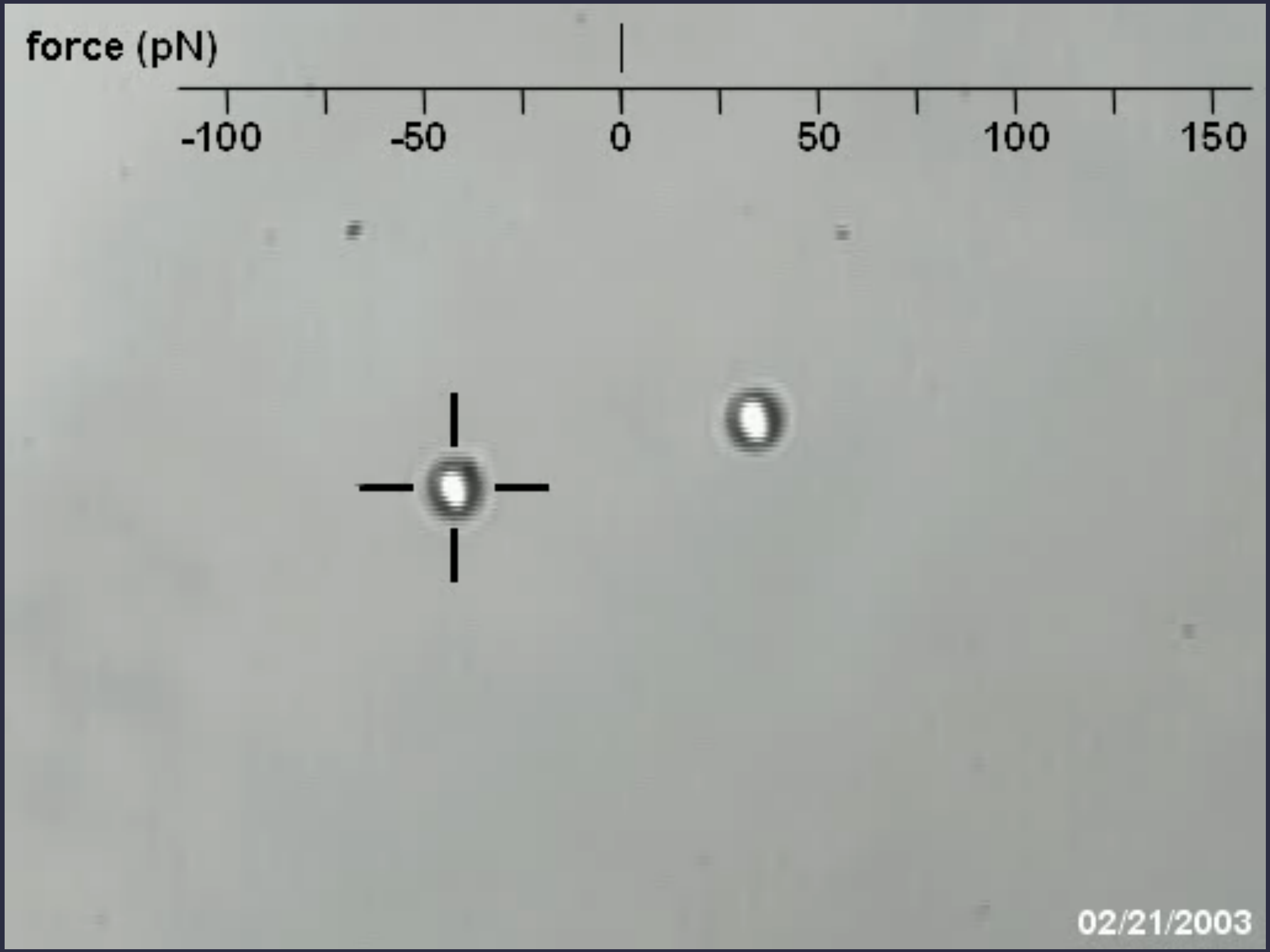


membrane of hair cell

inner ear hair cell



overstretching DNA with two optical traps

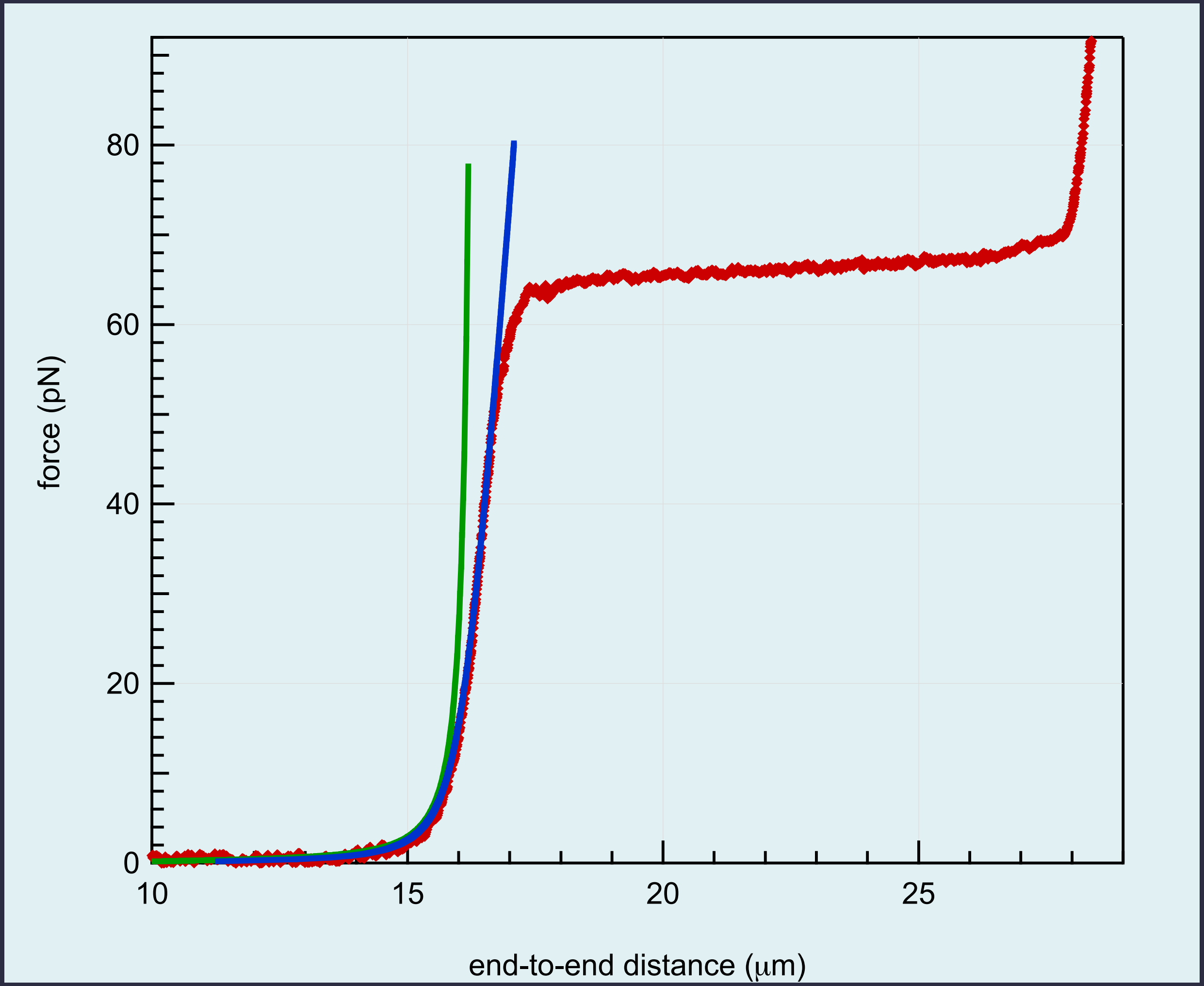


overstretching DNA with two optical traps

data

wormlike chain fit

stretchable wormlike chain fit



Optical Tweezers

Induced dipole:

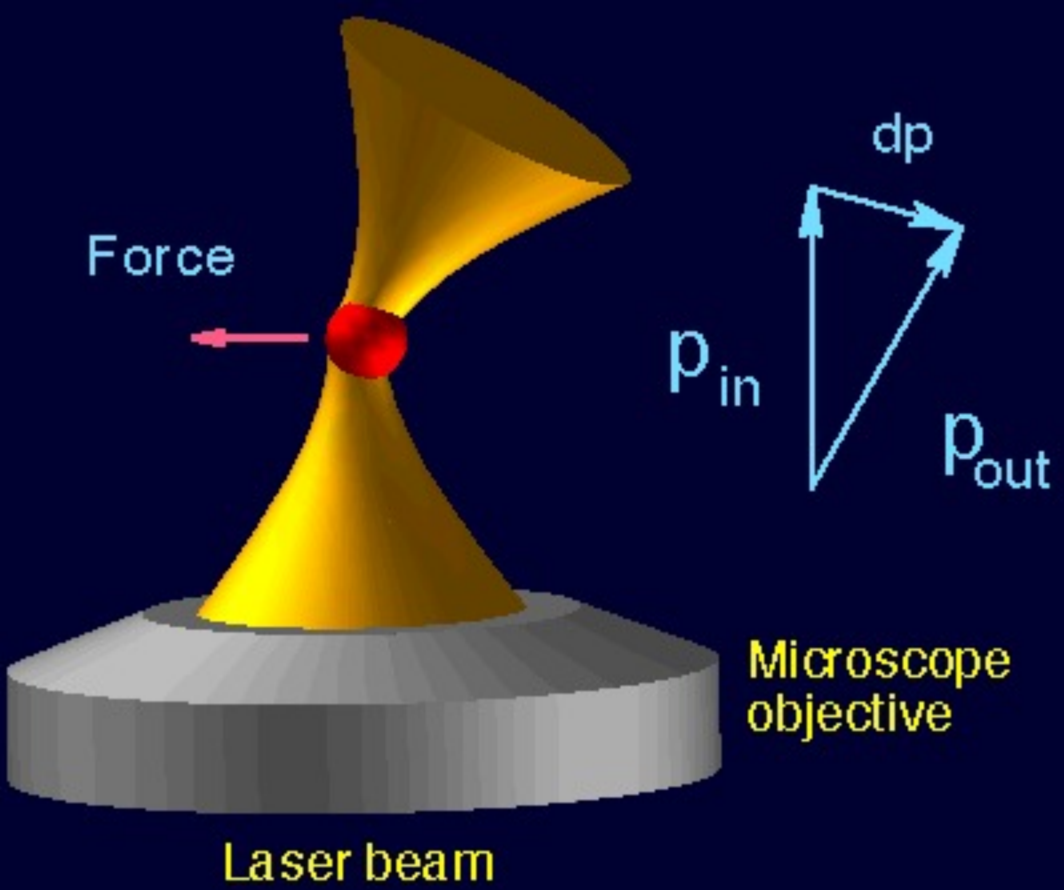
$\underline{d} = \alpha \underline{E}$

Energy:

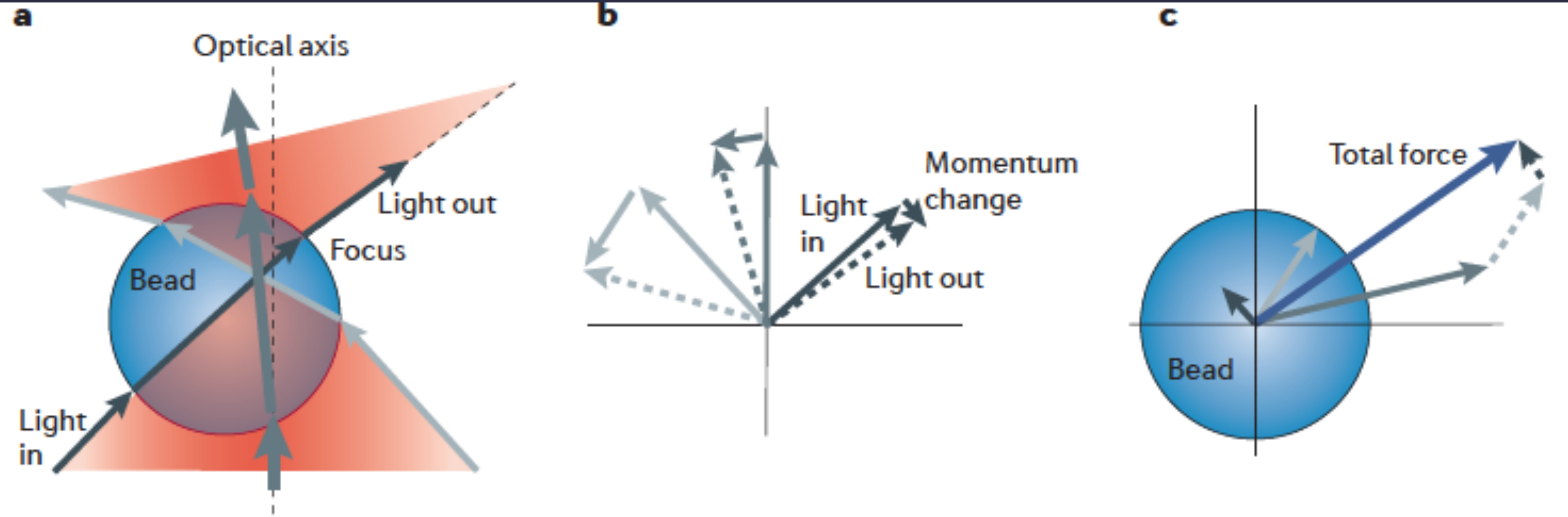
$U = -\underline{d} \circ \underline{E}$

Force:

$$\underline{F} = (\underline{d} \circ \nabla)\ \underline{E}$$
$$= \alpha/2\ \nabla (\underline{E})^2$$



geometrical optics limit: particle >> wave length

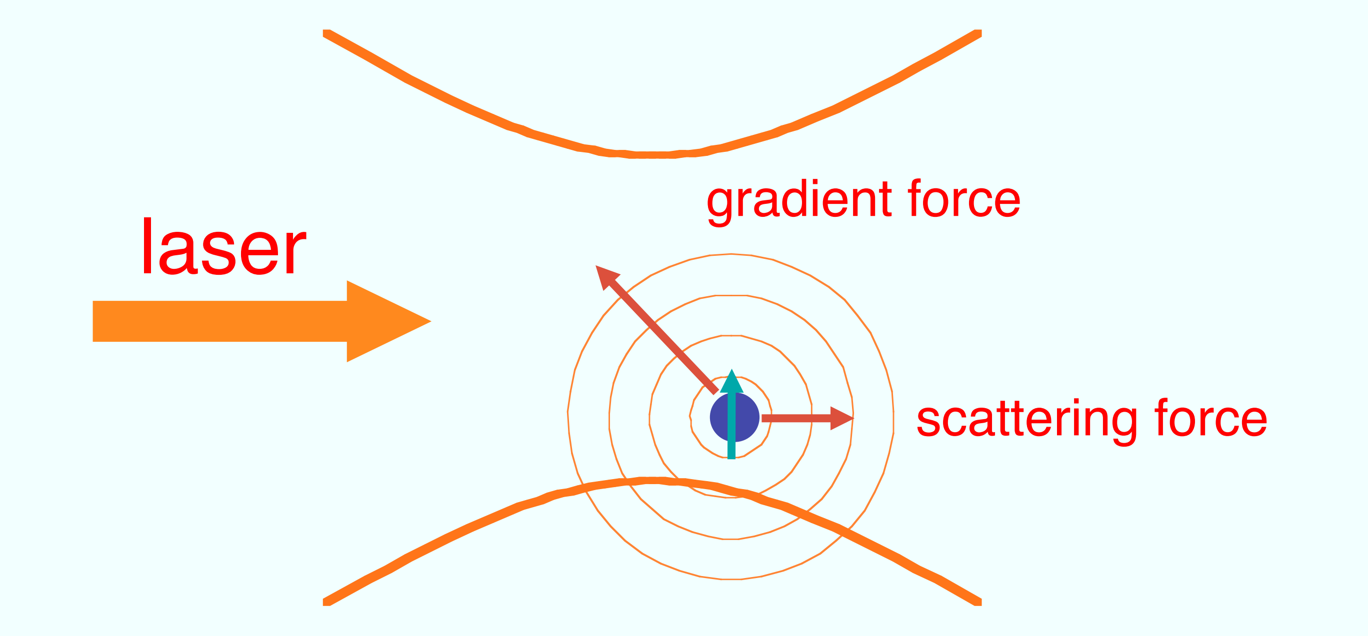


Refraction of light interacting with a bead near the focus of a laser beam

Momentum transfer on each photon

Total force on the bead

Rayleigh limit: particle << wave length



E ~ constant across the particle

some notes on trapability

- force on particle = momentum exchange with E-field

- 2 competing components: dissipative scattering force, conservative gradient force

$$\langle \mathbf{F_s} \rangle = \frac{\varepsilon_0}{2} \mathrm{Re}\left[-i\hat{\varepsilon}_j \alpha'' \mathbf{E_k^*} \cdot \left(\frac{\partial \mathbf{E_k}}{\partial x_j}\right) \right] \qquad \langle \mathbf{F_g} \rangle = \frac{\varepsilon_0}{2} \mathrm{Re}\left[\hat{\varepsilon}_j \alpha' \mathbf{E_k^*} \cdot \left(\frac{\partial \mathbf{E_k}}{\partial x_j}\right) \right]$$

- to get trap, need: $\langle \mathbf{F_g} \rangle > \langle \mathbf{F}_s \rangle$ (potential well > kT)

- polarizability of small sphere: $\alpha = (d/2)^3 \frac{n_r^2 - 1}{n_r^2 + 2}$ with relative index $n_r = \frac{n_{particle}}{n_{solvent}}$

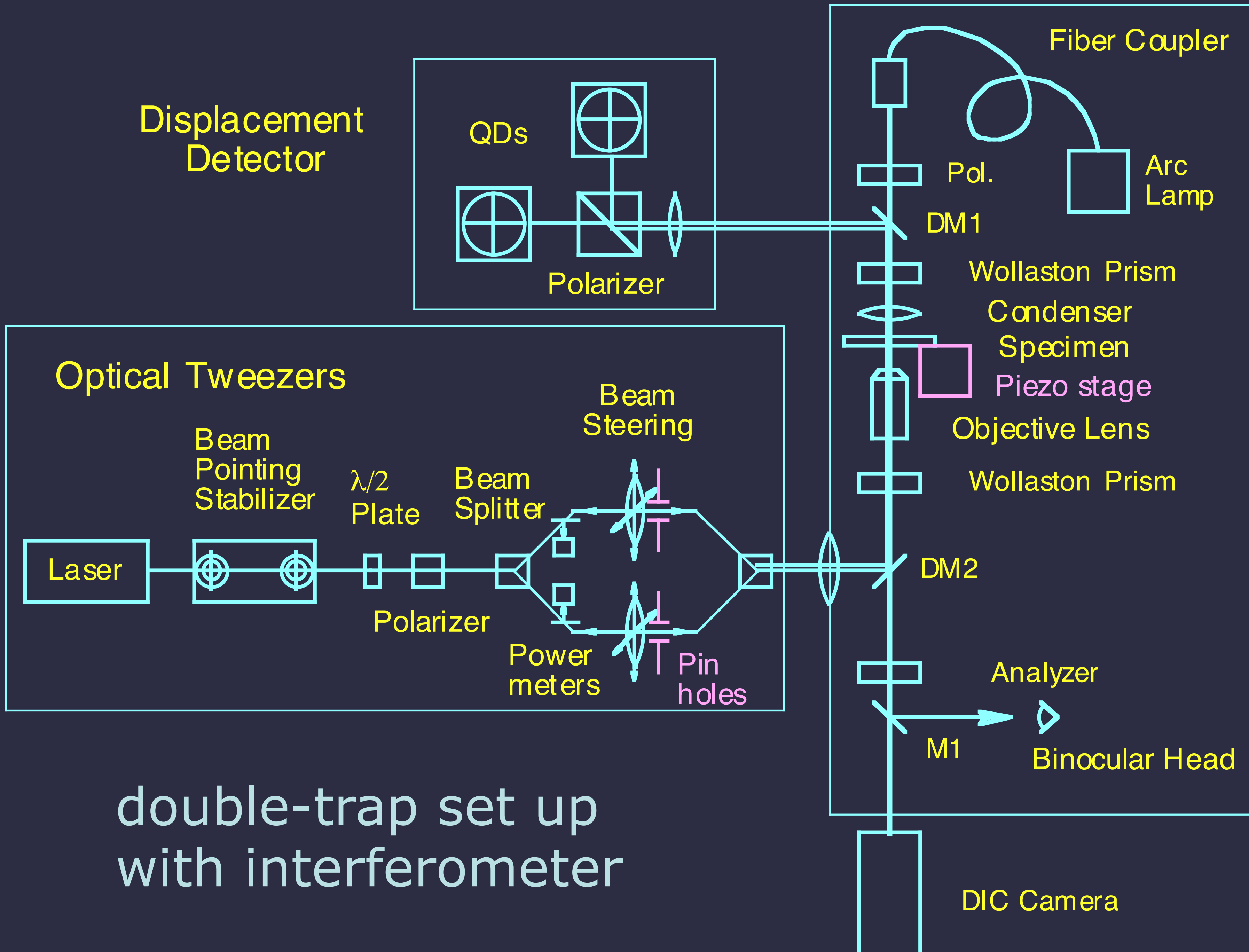
- even for non-absorbing particle: $\alpha'' \neq 0$ (radiation reaction),

but in power series expansion: $\alpha' \propto vol = d^3$ and $\alpha'' \propto vol^2 = d^6$

- this is related to Rayleigh scattering formula: $I_s \propto \frac{\alpha^2}{\lambda^4}$

- result: $\langle \mathbf{F}_s \rangle \propto \left(n_r d^3\right)^2$, $\langle \mathbf{F}_g \rangle \propto n_r d^3$ therefore: F_s wins for large particles and large n_r!!

note: for geometrical optics (particle > wavelength) -> F_g independent of size!



double-trap set up
with interferometer

lasers:

HeNe: 632 nm, ~1-30mW, usually a bit too weak, inexpensive

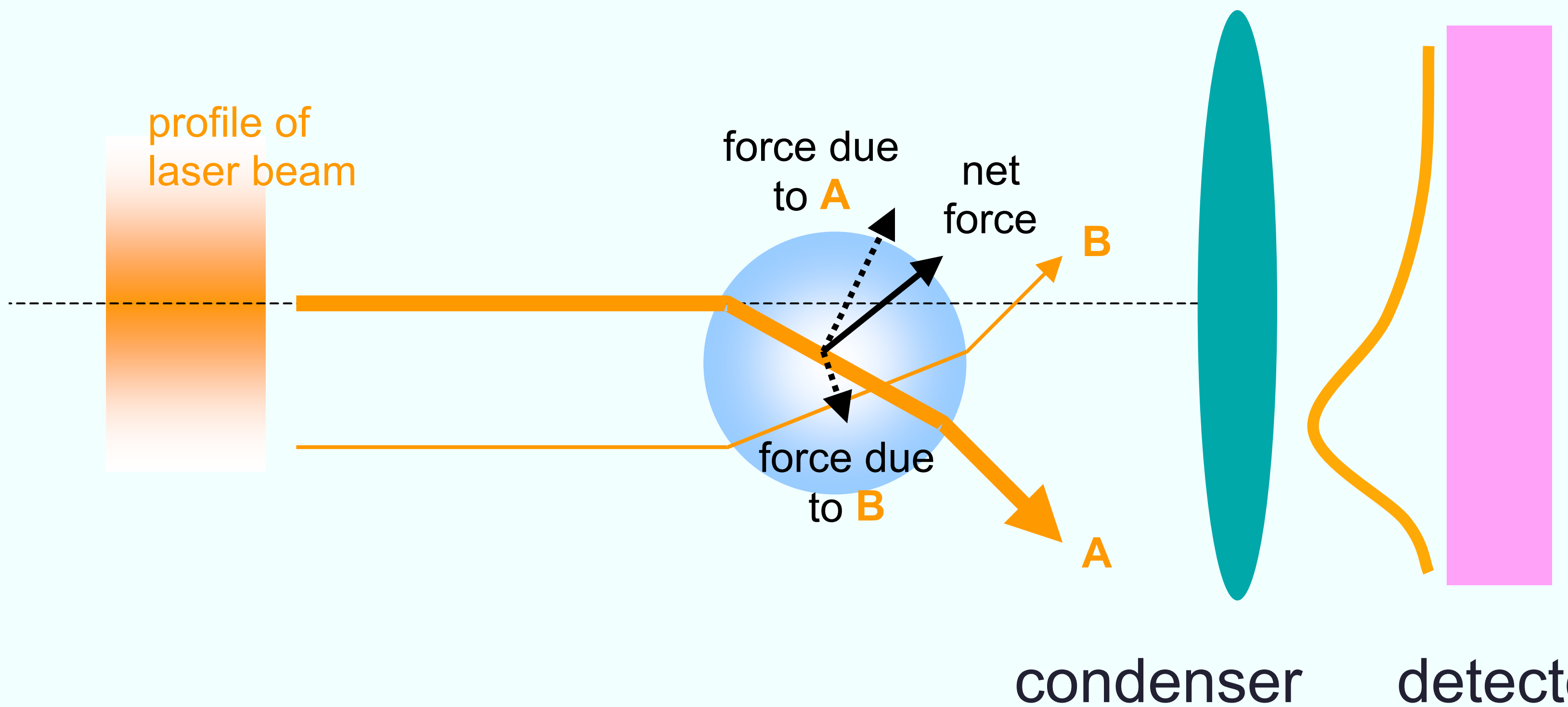
Diode lasers: blue to infrared, up to 100s of mW, inexpensive
note: need single-mode, beam profile not circular, need correction optics.

Ar-ion lasers: 488, 514nm, 100s of mW to 10s of W, expensive, water cooled, inefficient, outdated.

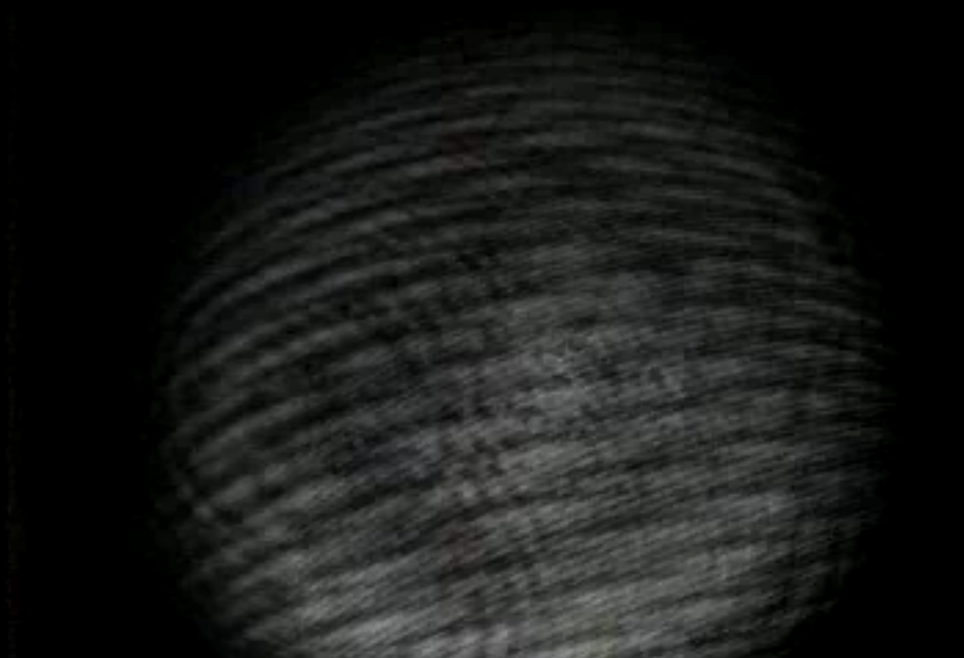
Rare earth, solid state lasers (diode pumped): Nd-YAG, ND-VO_4, ND-YLF, 1056-1064 nm, 100s of mW to 20W or more, standard for many purposes, not too cheap.

Various exotic designs: fiber laser, disk laser

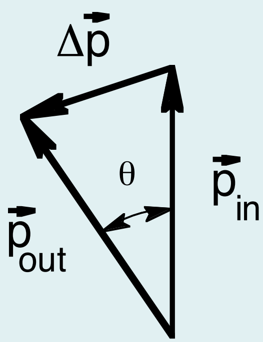
geometrical optics limit: particle >> wave length

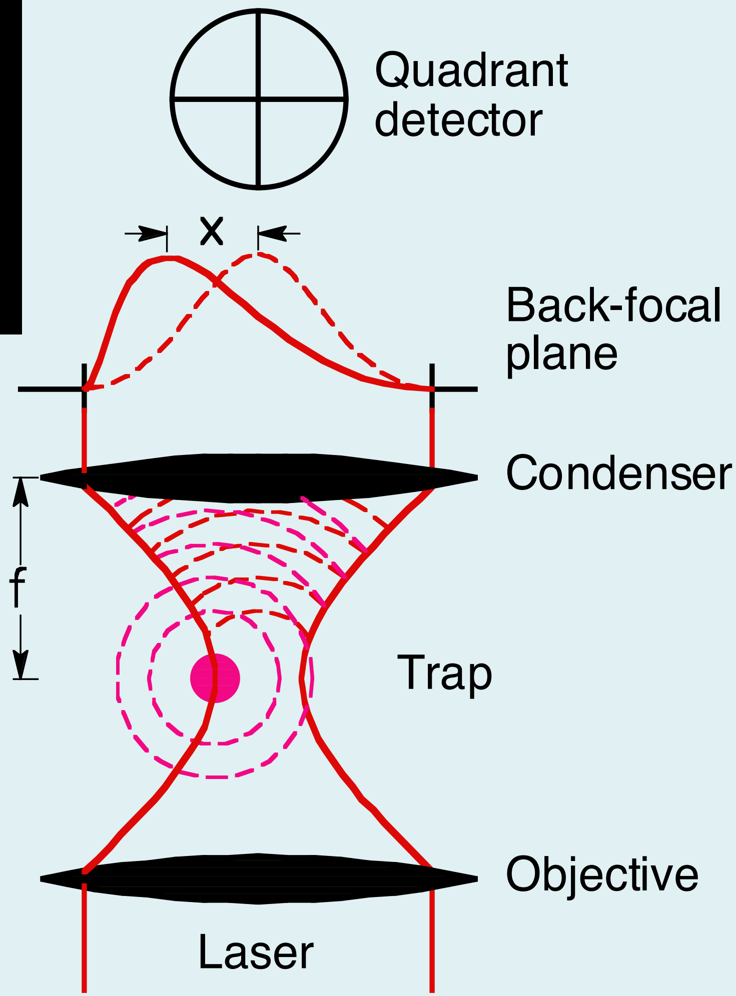


interferometric position detection



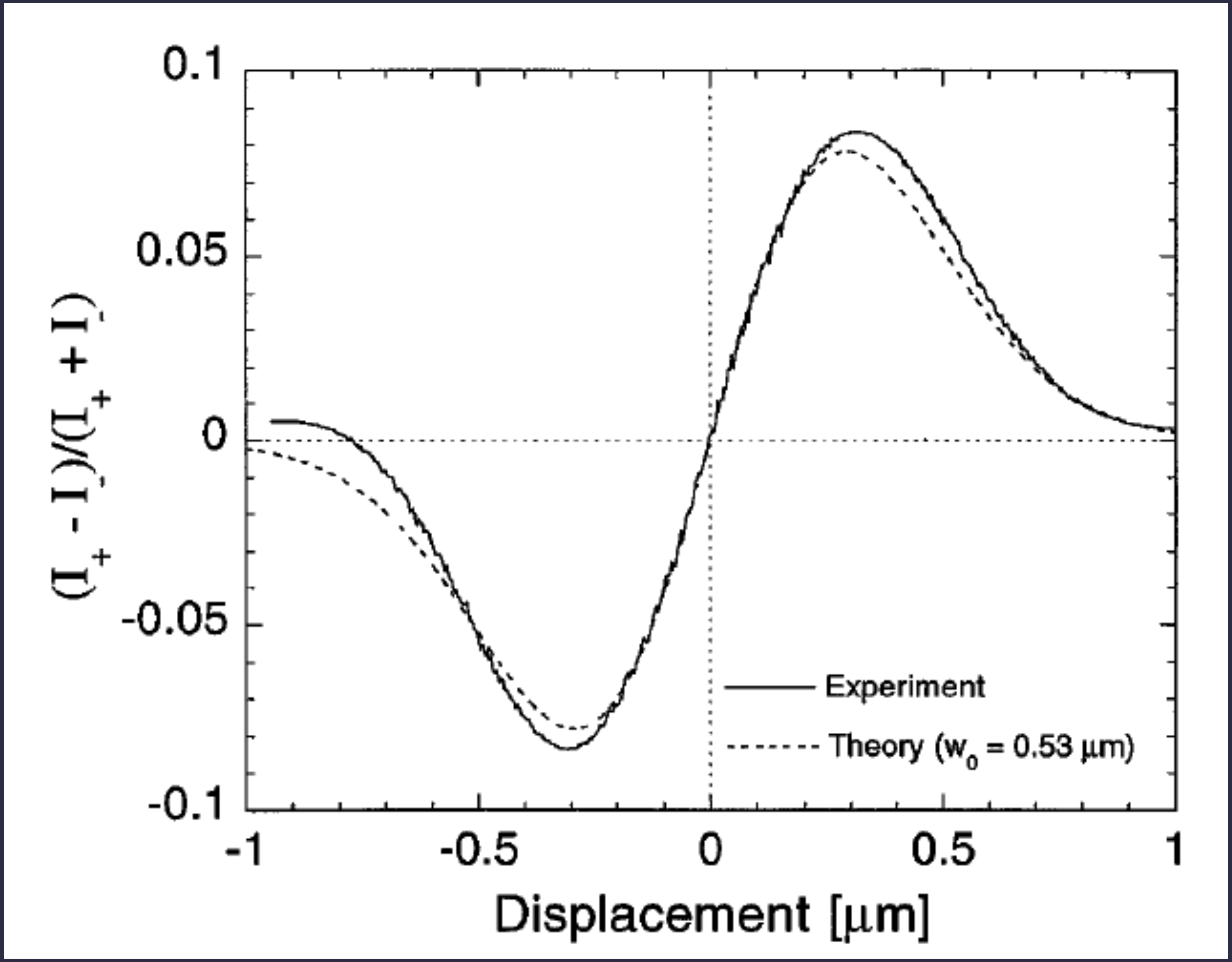
$$F = \frac{dp}{dt} = \frac{I}{c}\sin\theta = \frac{I}{c}\,\frac{x}{f}$$





Gittes, Schmidt, (1998), Opt. Lett. 23: 7-9.

comparison of first order interference model with data (no adjustable parameters)



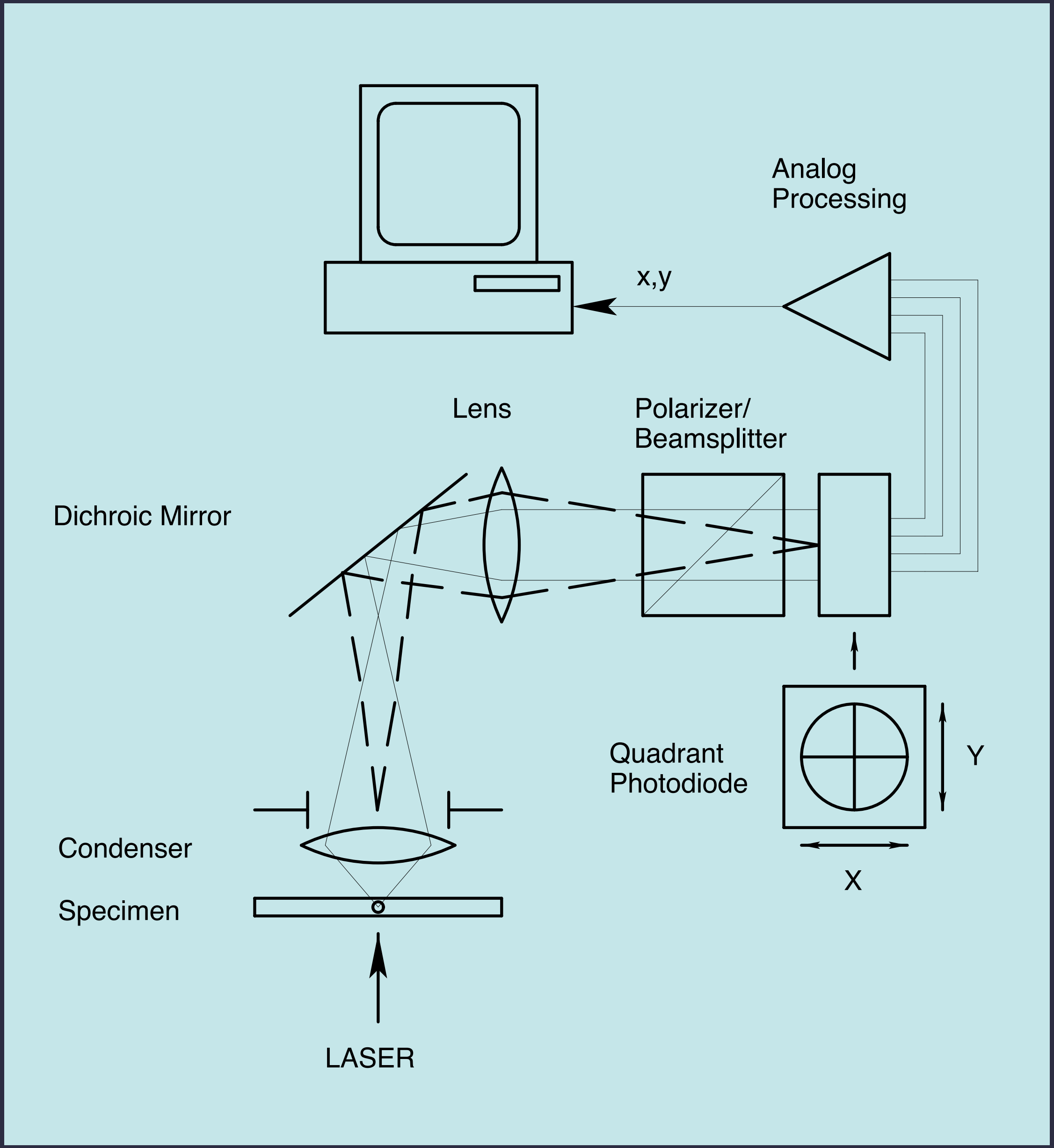
bead size: 0.5 μm

detectors

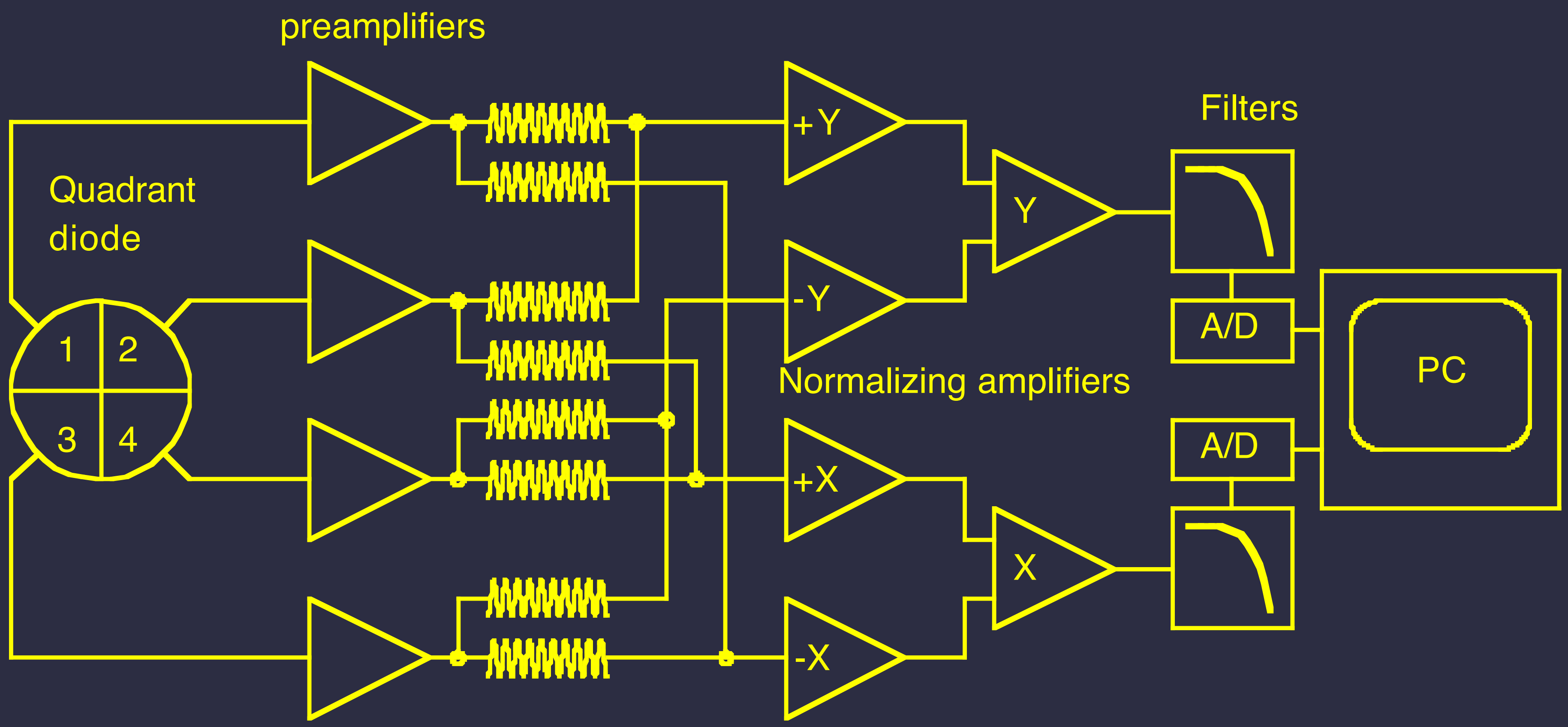
CCD camera

quadrant photodiodes

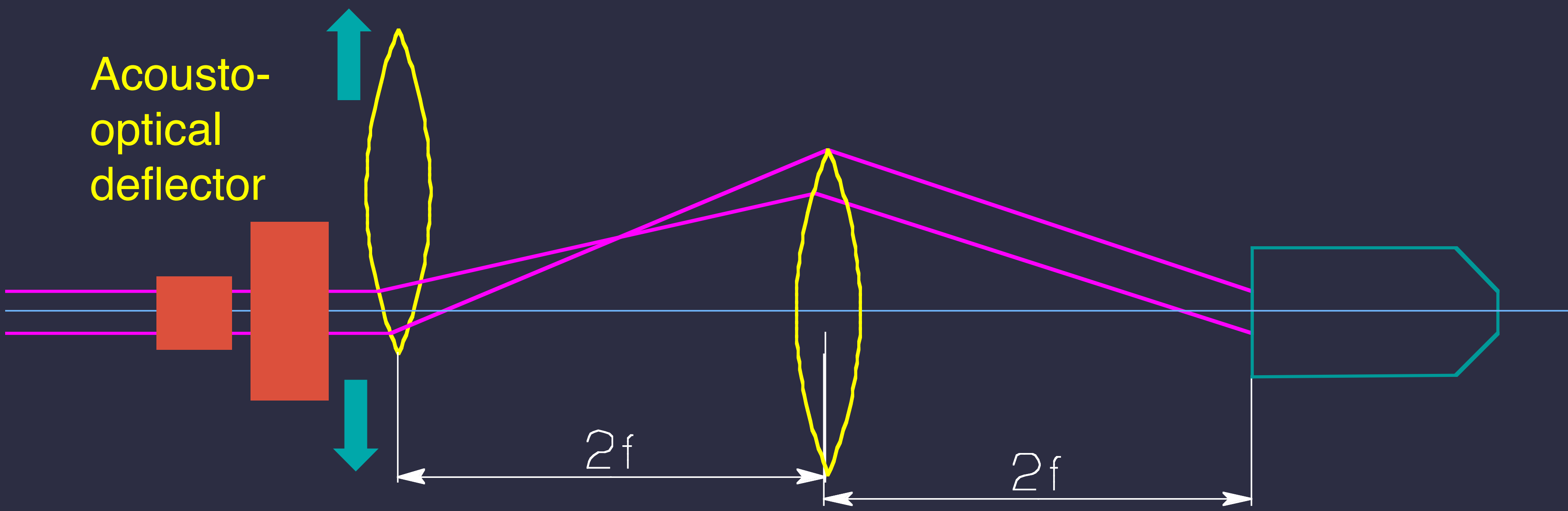
data processing



data processing:



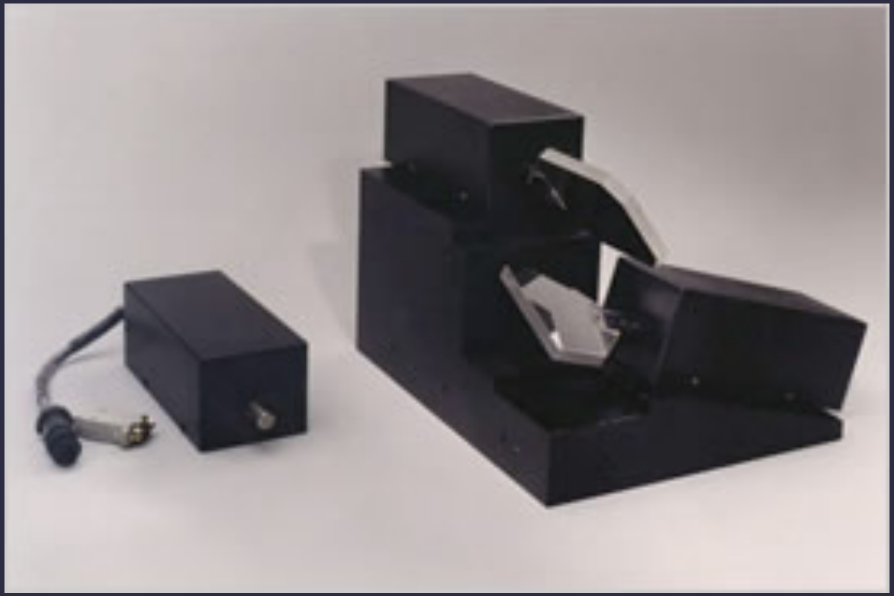
beam steering



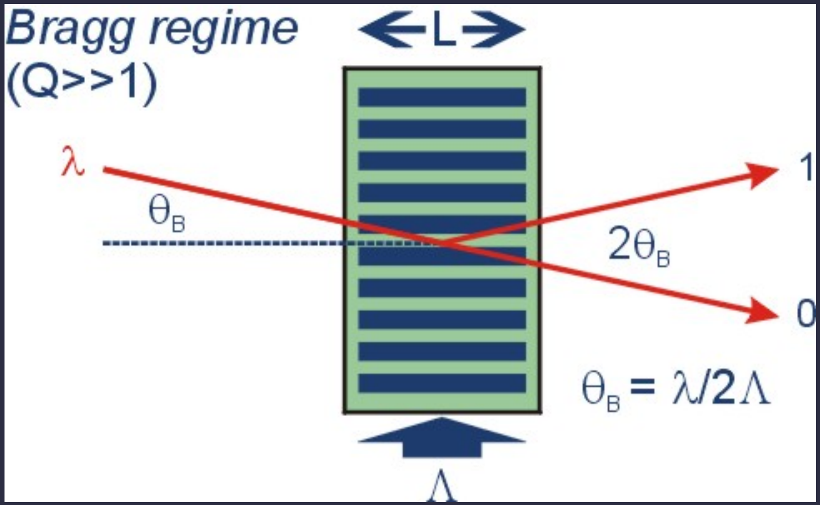
pivot the beam around point in entrance pupil of objective

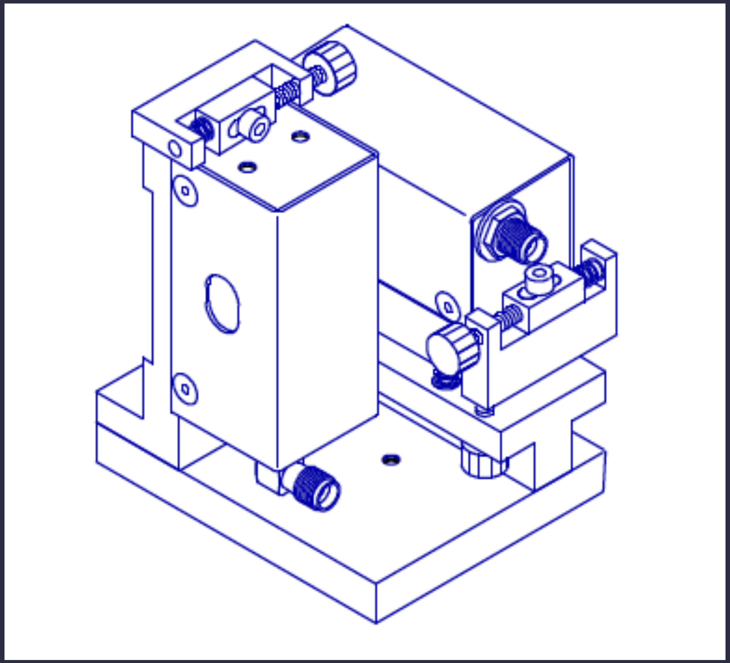
fast beam steering

Galvo-mirrors:

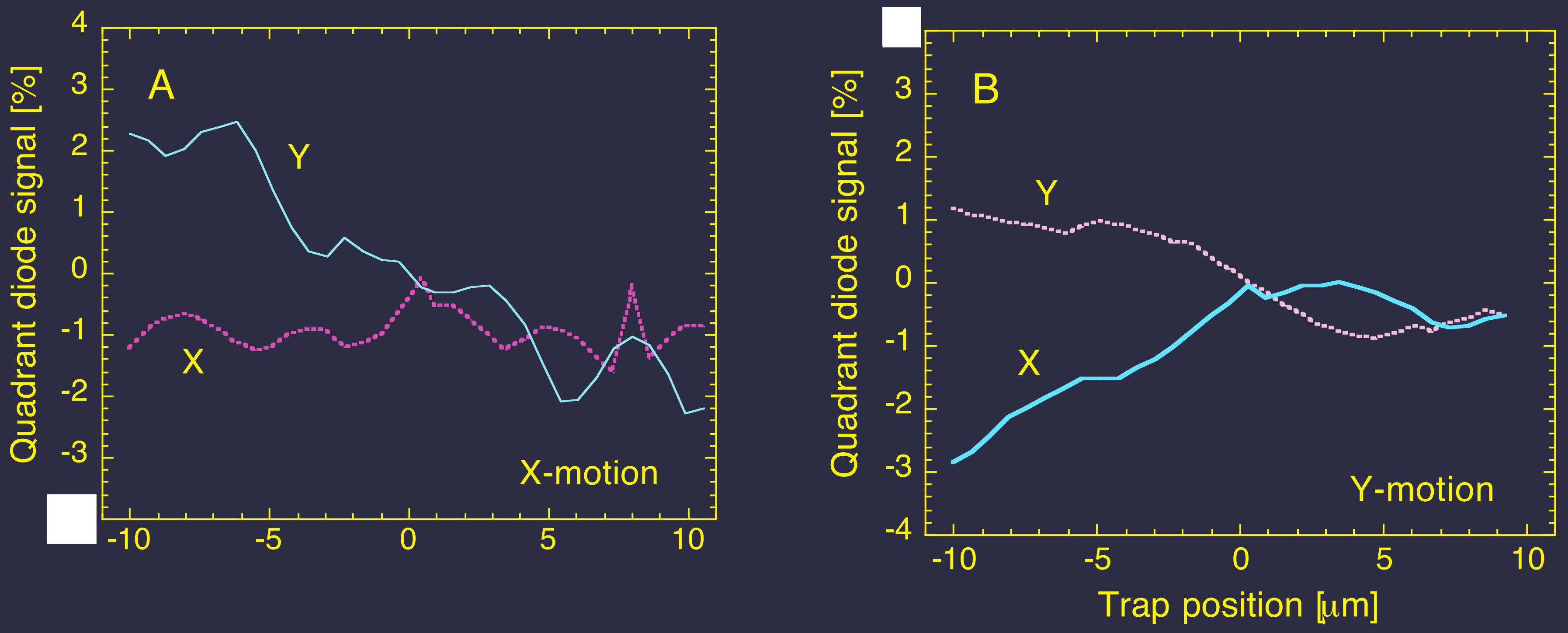


acousto-optic deflectors:
(tellurium oxide)



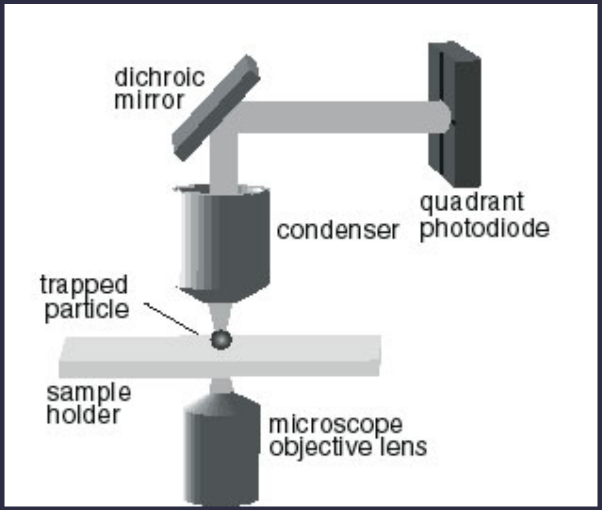


independence of trap position

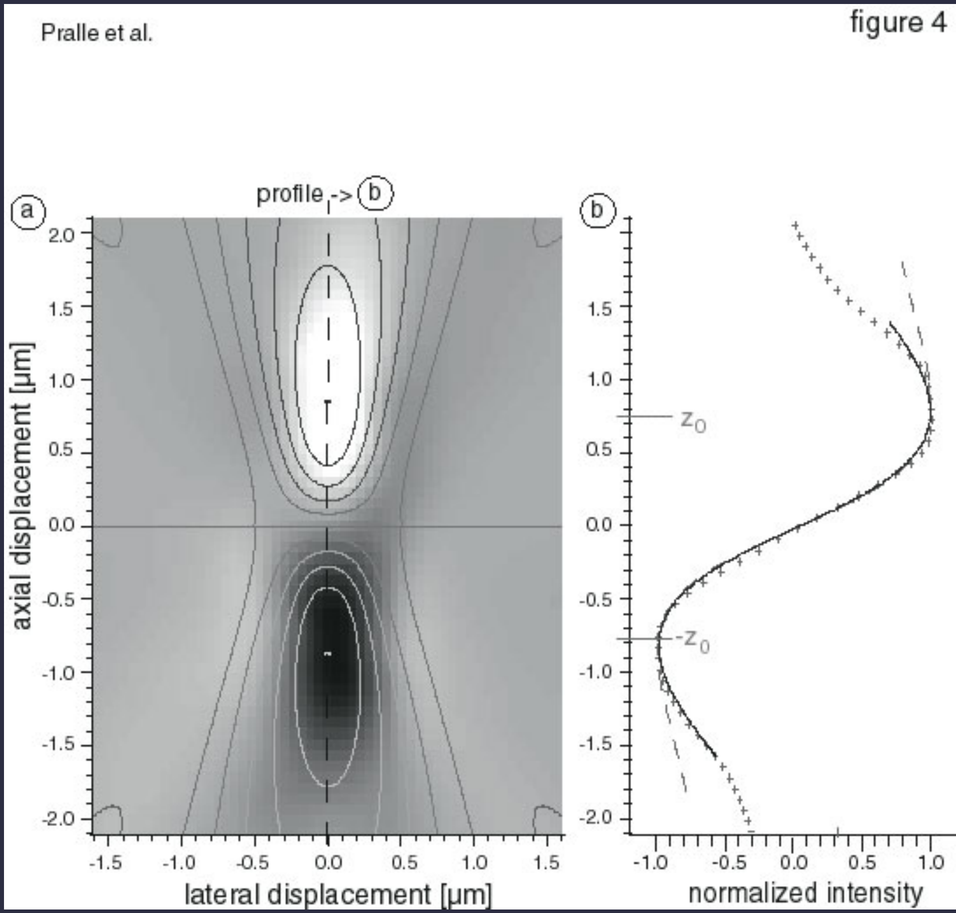


axial position detection

See: A. Pralle, M. Prummer, E.-L. Florin, E.H.K. Stelzer, J.K.H. Hoerber Microscopy Research and Technique 44:378 (1999)



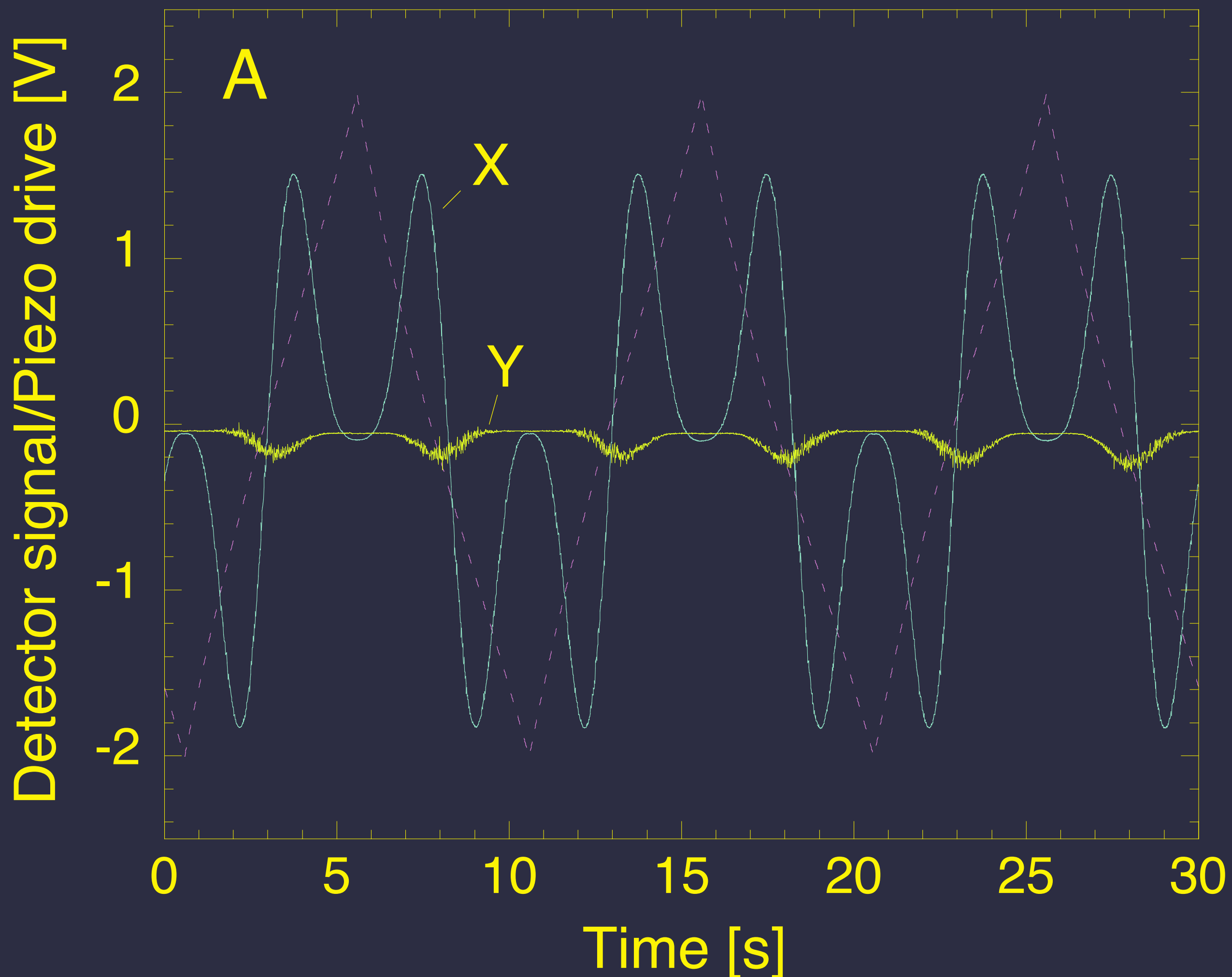
$$\frac{I_z}{I}(z') = \frac{8k\alpha}{\pi w_0^2}\left(1 + \left(\frac{z'}{z_0}\right)^2\right)^{-1/2} \sin\left(\arctan\frac{z'}{z_0}\right)$$



k: wave number
a: polarizability
w_o: Gaussian beam waist
z': axial displacement
z_0: Rayleigh range

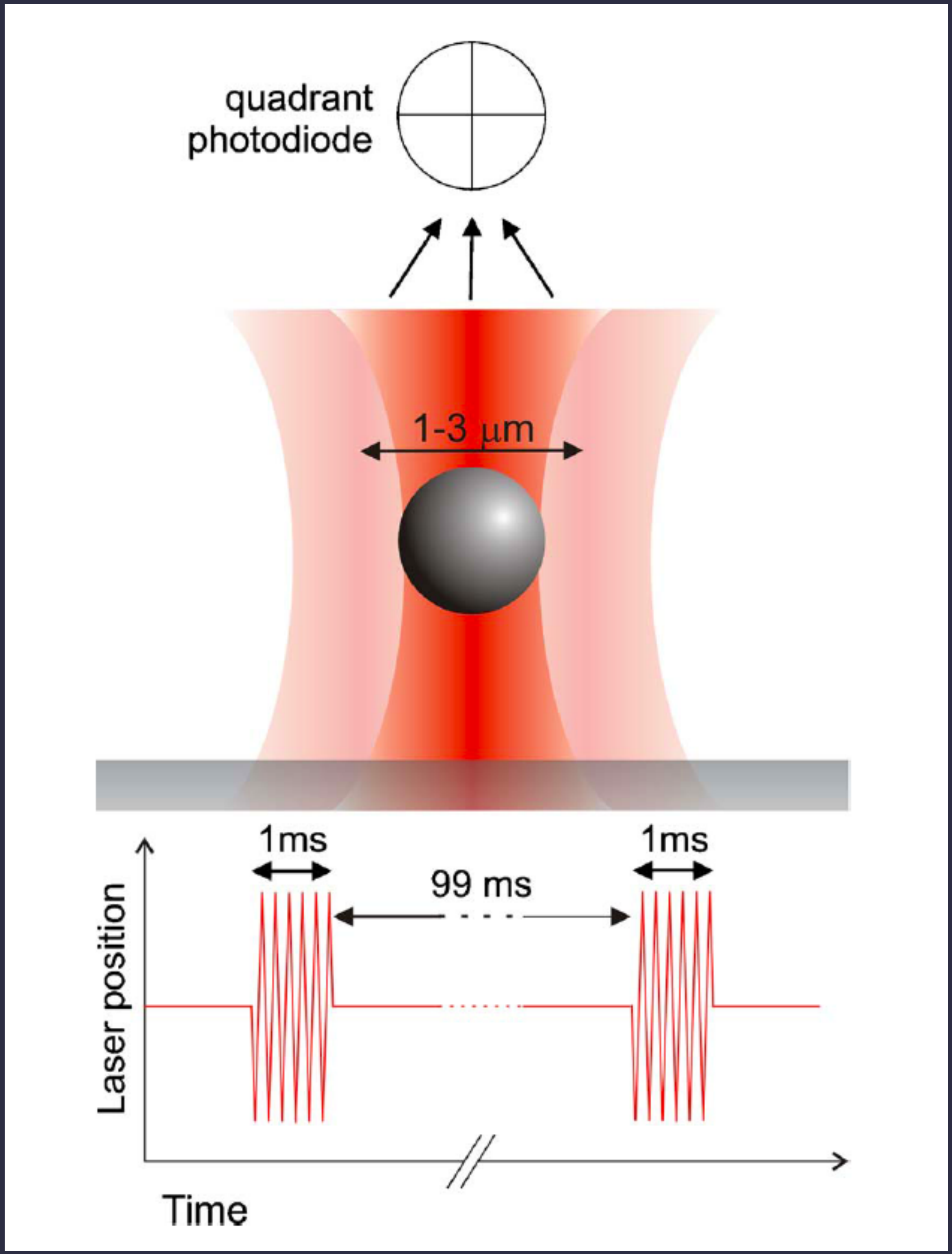
calibration of position detection

direct method: with piezo stage

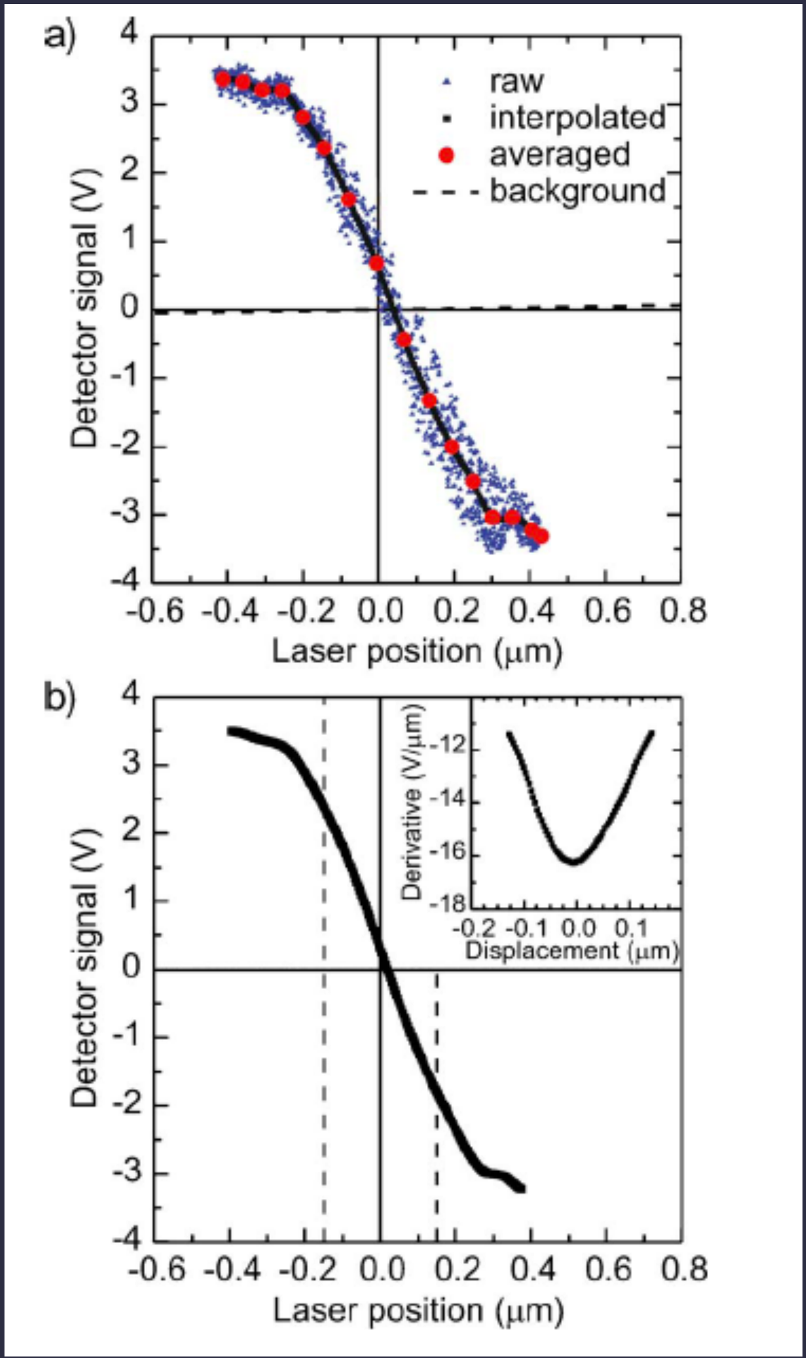


“wiggle method”

- no need to fix bead,
- done on the same bead as used later,
- can be done in media of unknown viscoelasticity



detector response around zero displacement



microscope objective transmission curves

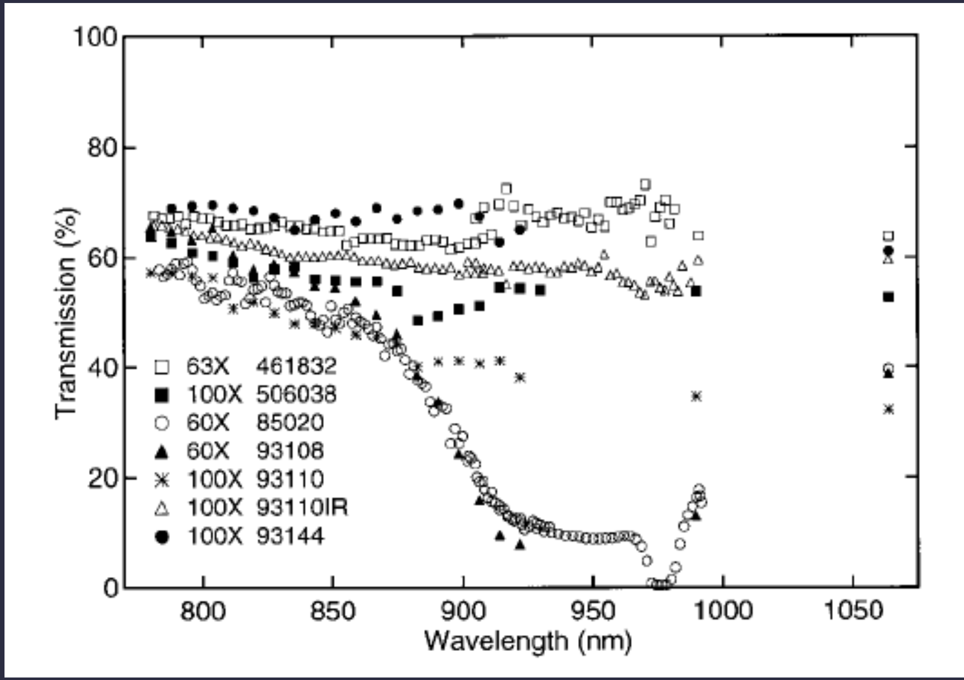


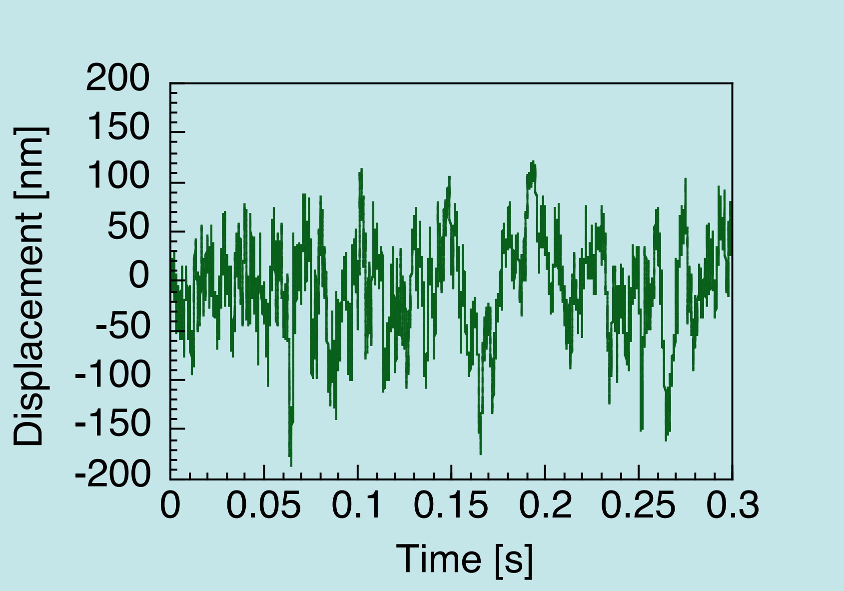
TABLE 1 Transmission of microscope objectives, cross-referenced with Fig. 2

Part no.	Manufacturer	Magnification/tube length (mm)/numerical aperture	Type designation	Transmission (±5%) 830 nm	850 nm	990 nm	1064 nm
461832	Zeiss	63/160/1.2 water	Plan NeoFluar	66	65	64	64
506038	Leica	100/∞/1.4–0.7 oil	Plan Apo	58	56	54	53
85020	Nikon	60/160/1.4 oil	Plan Apo	54	51	17	40
93108	Nikon	60/∞/1.4 oil	Plan Apo CFI	59	54	13	39
93110	Nikon	100/∞/1.4 oil	Plan Apo CFI	50	47	35	32
93110IR	Nikon	100/∞/1.4 oil	Plan Apo IR CFI	61	60	59	59
93144	Nikon	100/∞/1.3 oil	Plan Fluor CFI	67	68	—	61

K. C. Neuman et al. , Biophys. J. 77: 2856 (1999)

Calibration from fluctuation data in viscous solution

Discretely sampled positions



Fourier transform:

$$X(f_m) = \sum_{n=1}^{N} x_n e^{2\pi i n m / N}$$

Frequency/resolution:

$$f_m = m\,\delta f \qquad \delta f = \frac{1}{N\delta t}$$

Position power-spectral Density:

$$S(f_m) = \frac{2}{N^2 \delta f} \left| X(f_m) \right|^2$$

Langevin eq. for trapped bead:

$$\gamma \frac{dx}{dt} + \kappa x = F(t)$$

Thermal force:

$$\langle F(t) \rangle = 0 \quad S_F(f) = \left| F(f) \right|^2 = 4\gamma k_b T$$

FT position

$$x(t) = \int_{-\infty}^{+\infty} X(f) e^{-2\pi i f t} df$$

FT of dx/dt:

$$-2\pi i f X(f)$$

FT of Langevin eq.

$$2\pi\gamma(f_c - if)X(f) = F(f)$$

Corner frequency:

$$f_c = \frac{\kappa}{2\pi\gamma}$$

Define position PSD:

$$S_x(f) = \left| X(f) \right|^2$$

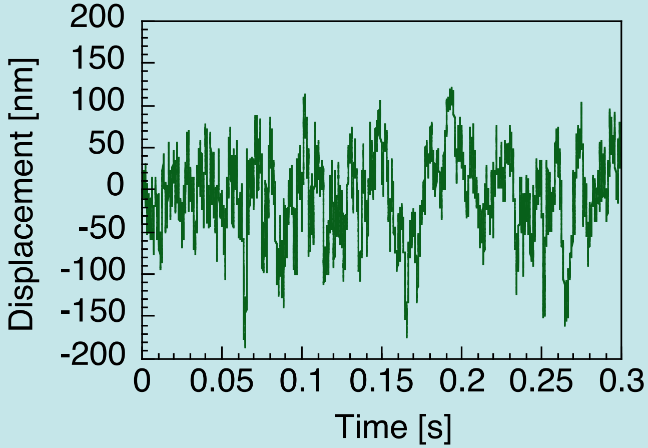
Take squared modulus:

$$4\pi^2\gamma^2\left(f_c^{\,2} + f^{\,2}\right)S_x(f) = S_F(f)$$

Position PSD:

$$S_x(f) = \frac{k_b T}{\gamma\pi^2\left(f_c^{\,2} + f^{\,2}\right)}$$

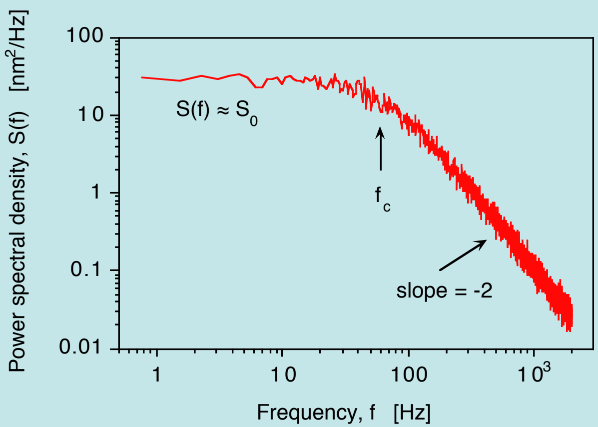
Thermal Motion of a Trapped/Tethered Particle



Time series:

$$\mathrm{var}(x) = \langle x^2 \rangle - \langle x \rangle^2 = \frac{k_B T}{\kappa}$$

(equipartition)



Spectrum:

$$S(f) = \frac{k_B T}{\pi^2 \gamma (f_c^2 + f^2)}$$

$$f_c = \frac{\kappa}{2\pi\gamma}, \qquad S_0 = \frac{4\gamma k_B T}{\kappa^2}$$

trapped bead attached to motor: $\mathrm{var}(x) = \dfrac{k_B T}{\kappa_{trap} + \kappa_{motor}}$

See: Gittes & Schmidt, 1998, Meth. Cell Biol. 55:129

Calibration using the spectral density of Brownian motion

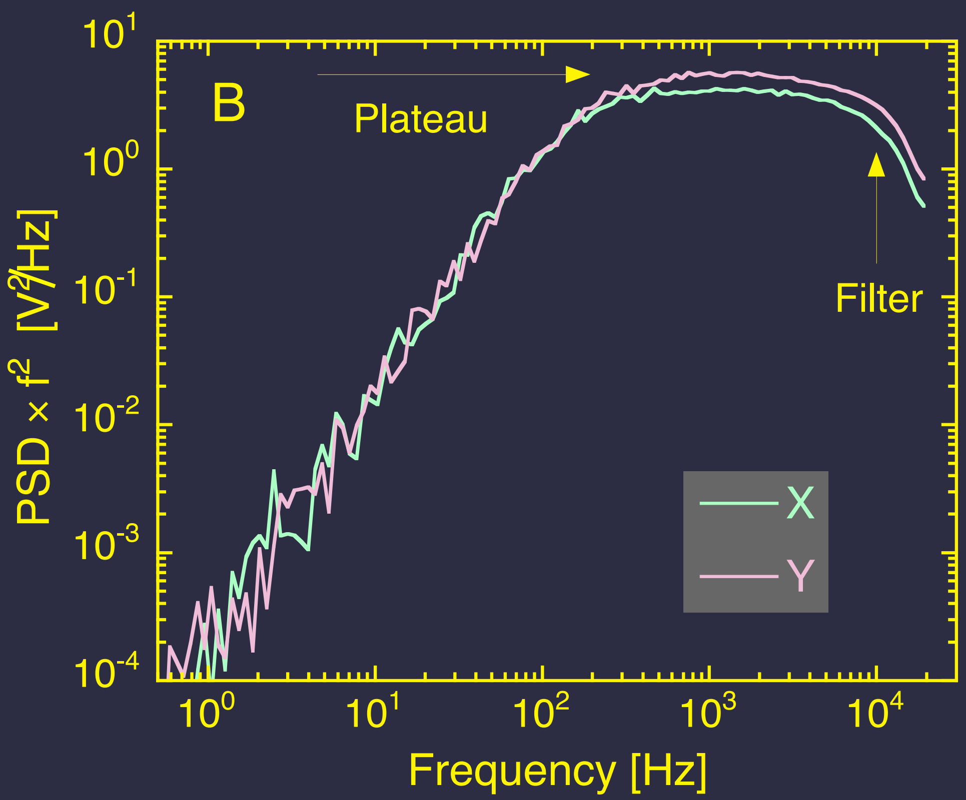
Brownian bead in trap:

PSD: $$S(f) = \frac{S_0 f_0^2}{f_0^2 + f^2}$$

intercept: $$S_0 = 4\gamma k_b T / \kappa^2$$

corner frequency: $$f_0 = \frac{\kappa}{2\pi\gamma}$$

drag: $$\gamma = 3\pi\eta d$$

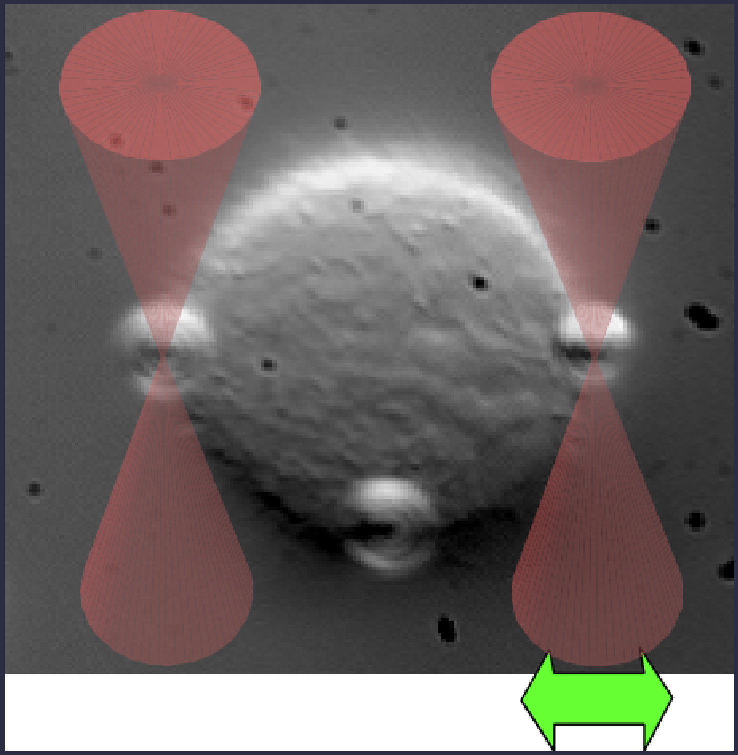


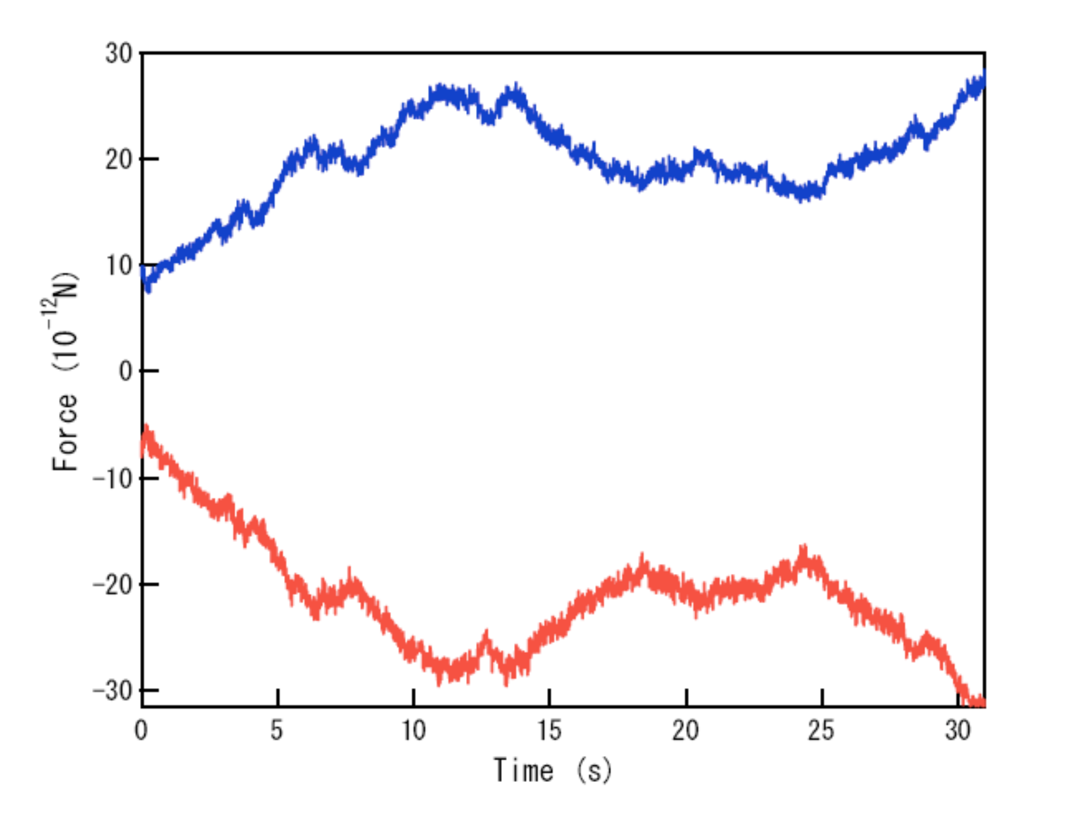
PSD in Volts $$S^V(f) = \beta^2 S(f)$$

High-f plateau $$P^V = \beta^2 S_0 f_0^2$$

Calibration factor $$\beta = \sqrt{\frac{3\pi^3 \eta P^V d}{k_b T}}$$

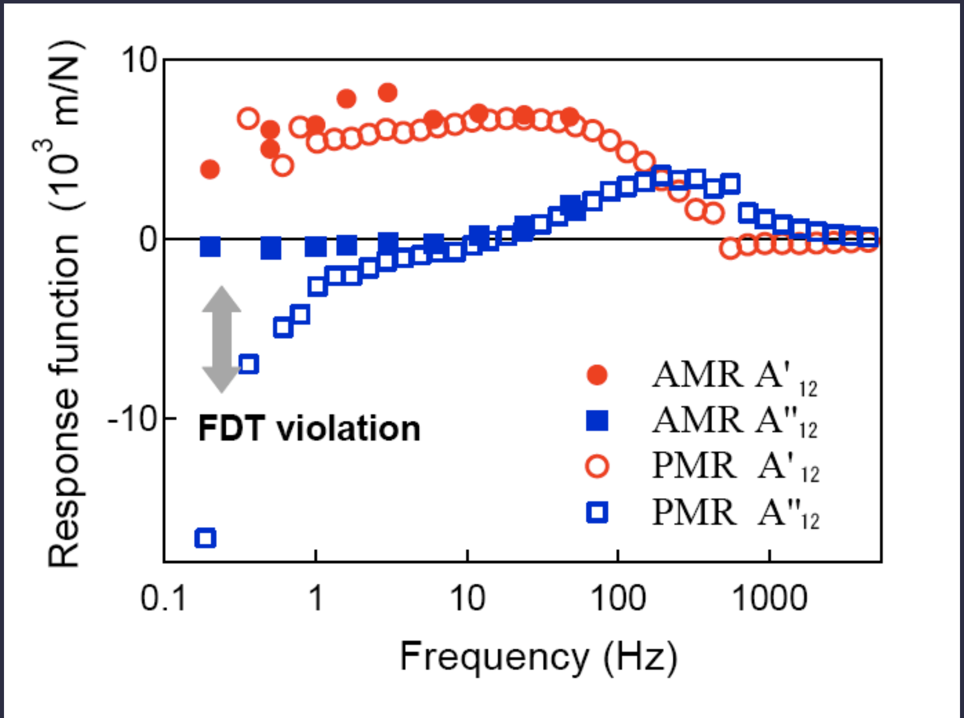
anti-correlated fluctuations of a trapped cell



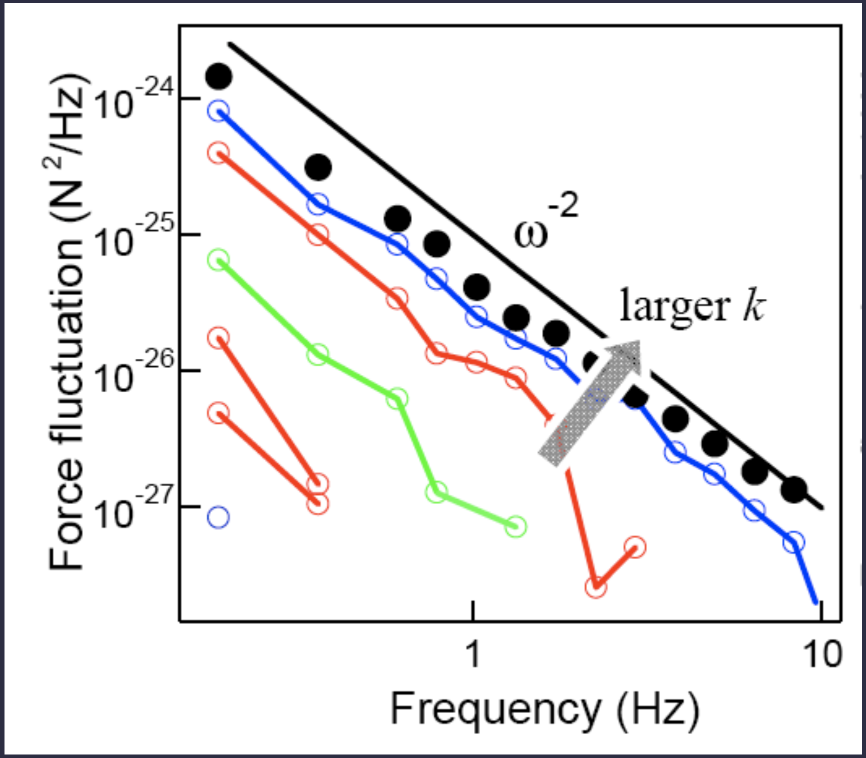


spectrum of anti-correlated fluctuations

response function



force fluctuation spectrum



the optical spanner: spin and orbital angular momentum of a laser beam

Wave equation:

$$\nabla^2 u(r,t) - \frac{1}{c^2}\frac{\partial^2 u(r,t)}{\partial t^2} = 0$$

Hermite-Gaussian:

$$u_{l,m}(x,y,z) = A_{l,m}\frac{w_0}{w(z)} H_l\left(\frac{x\sqrt{2}}{w(z)}\right) H_m\left(\frac{y\sqrt{2}}{w(z)}\right) e^{-\frac{x^2+y^2}{w^2(z)}} e^{-ikz} e^{-ik\frac{x^2+y^2}{2R(z)}} e^{i(l+m+1)\varphi}$$

Laguerre-Gaussian:

$$u_{l,m}(r,\varphi,z) = \frac{C}{\sqrt{1+z^2/z_r^{\,2}}}\left(\frac{r\sqrt{2}}{w(z)}\right)^m L_l^m\left(\frac{2r^2}{w^2(z)}\right) e^{-\frac{r^2}{w^2(z)}} e^{-\frac{ikr^2 z}{2\left(z^2+z_r^{\,2}\right)}} e^{-im\varphi} e^{i(2l+m+1)\tan^{-1}(z/z_r)}$$

Spin (linear, circularly polarized):

$$\sigma_z = 0, \pm 1$$

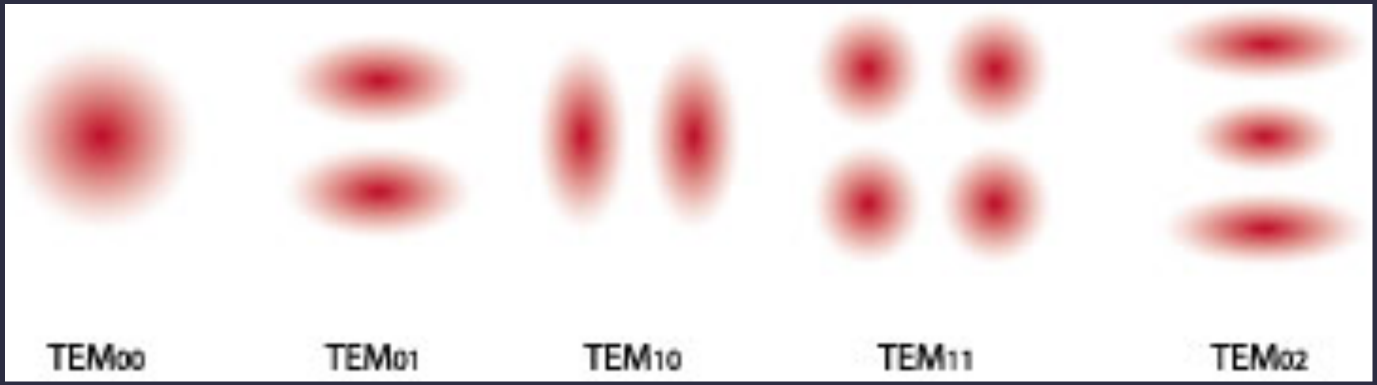
Total angular momentum, exact:

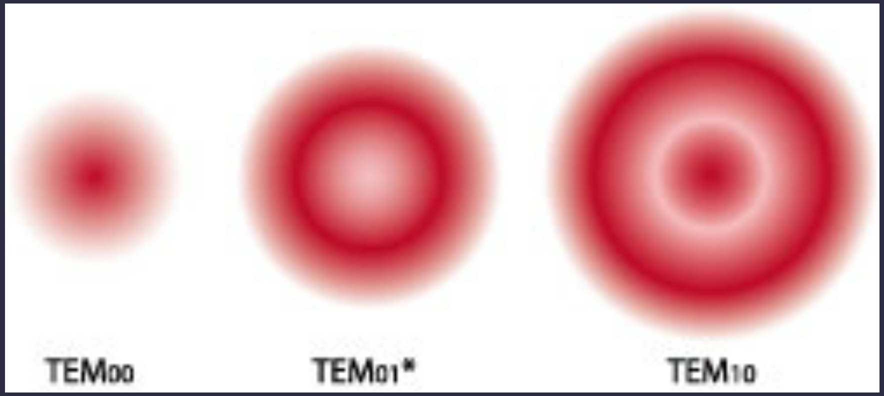
$$\left[l + \sigma_z + \sigma_z\left(\frac{2kz_r}{2p+l+1}+1\right)^{-1}\right]\hbar$$

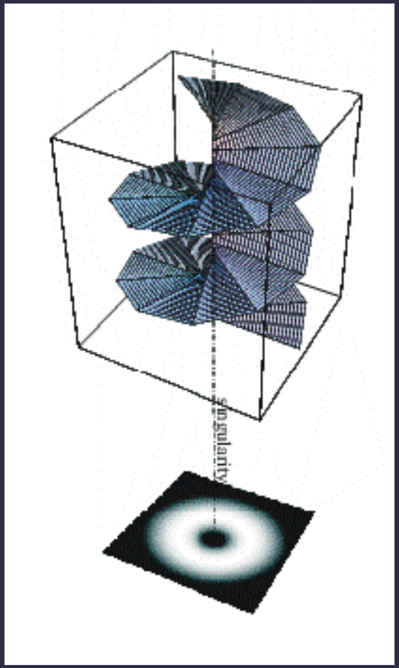
Collimated beam:

$$kz_r >> 1 \;\; \text{->} \;\; (l+\sigma_z)\hbar$$

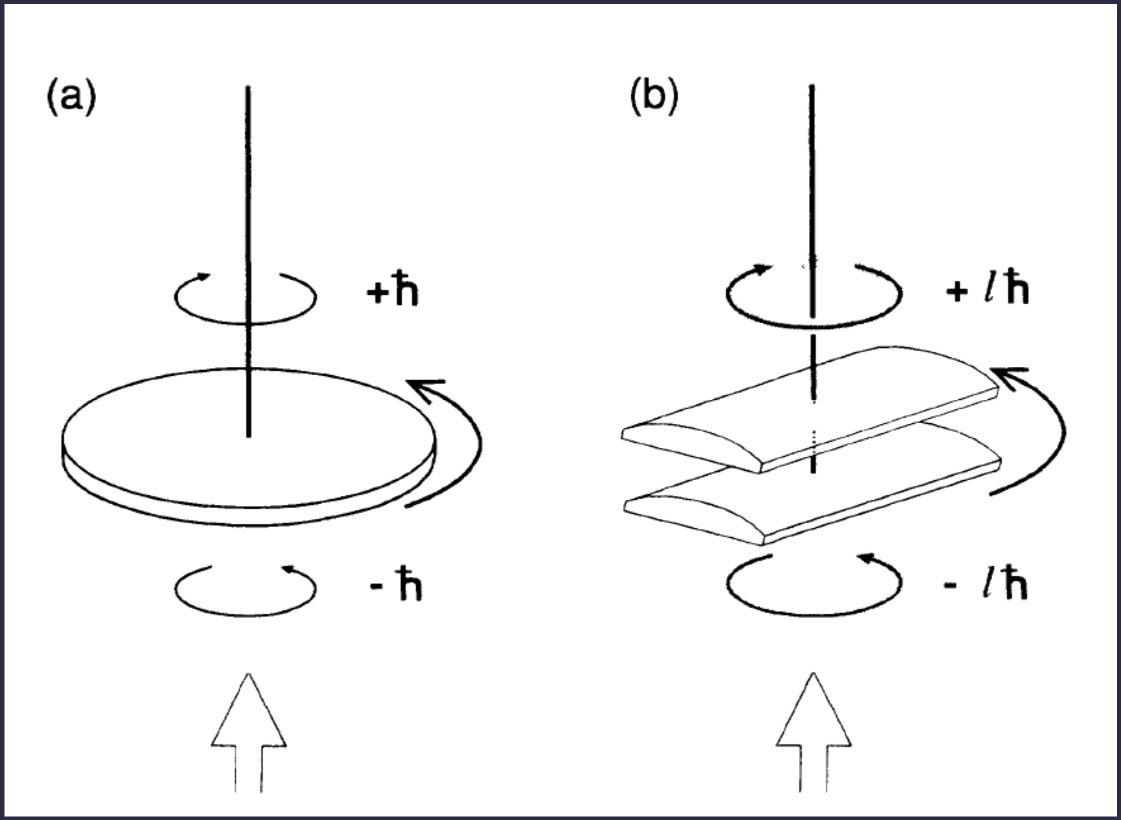
laser modes: Hermite-Gaussian, Laguerre-Gaussian



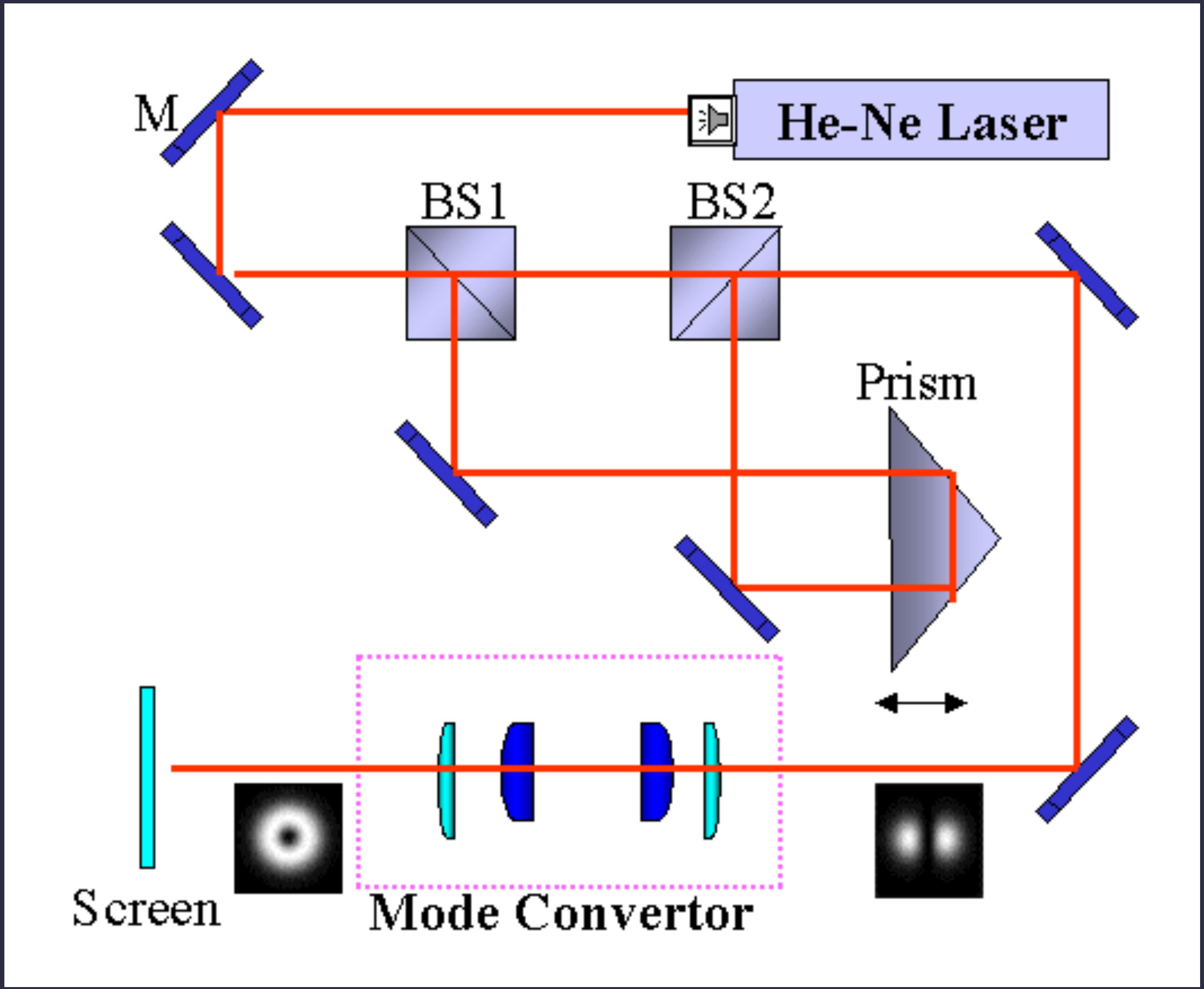




torque on optical elements changing
spin and orbital angular momentum of a laser beam



creating an optical spanner



Example:

Particle: teflon, 1 μm absorbing~2% at 1047 nm

power ~ 25 mW

L = 1 LG mode

f ~ 1Hz

with: $\tau = 8\pi\eta r^3\omega$

$\tau = 10^{-20}$ Nm = 0.1 pN nm

Simpson et al.
Opt. Lett 22:52 (1997)

THE END

Tetris produced by:
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