



# Optical Trapping and Force Measurement



## The Optical Trap

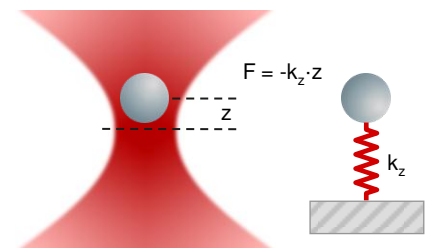
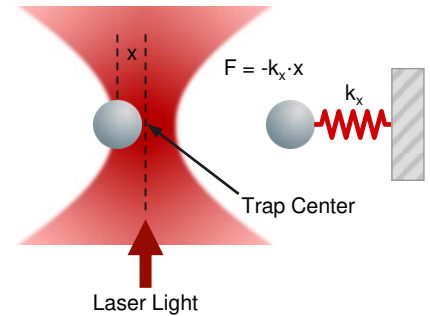
Microscopic objects - like individual nano- or microparticles, cells, cell compartments, single or clustered molecules - can be trapped securely inside the center of a strongly focused laser beam.

When an external force is acting on the trapped object, it deflects from the center of the trap. The deflection depends linearly on trap stiffness  $k$  and force  $F$ .

## Forces

Any trapped object experiences various external forces: Atoms or molecules of the surrounding medium induce Brownian motion in all three dimensions, depending on temperature, viscosity and the presence of obstacles in proximity. Macroscopic fluid movements cause drag forces. Electric fields, bulk or surface charges may generate electrophoretic or electroosmotic forces.

Particularly, single molecules bound to the trapped object can induce forces of broad variety and magnitude. The application of a force generated by an optical trap to a single molecule will gain vast insights into its molecular structure and elasticity, binding properties and kinetics.

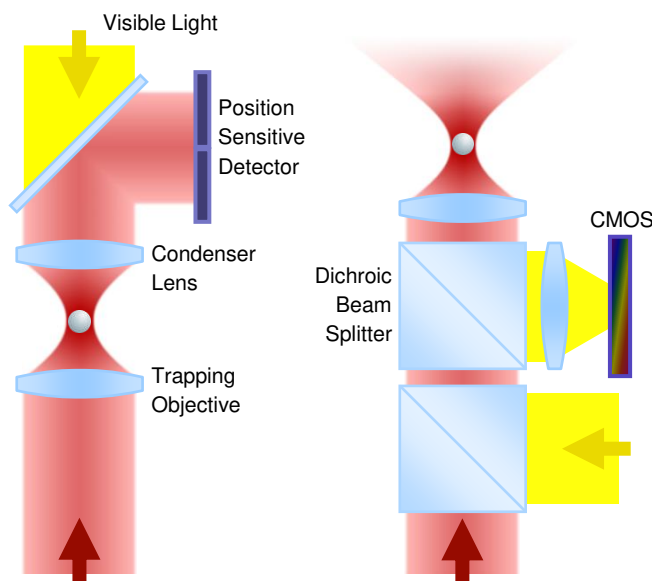


Lateral and axial forces acting on a trapped object.

## Deflection is the Essence

Generating and metering various forces requires a reliable force measurement capability in all three dimensions to allow for a maximum degree of experimental versatility and freedom. Thus, force detection must be accomplished by precisely measuring the deflection of the trapped particle in each direction.

**PicoTweezers** utilizes a sophisticated and easy-to-use video analysis for particle detection, tracking and force measurements. It provides the largest field of application since it avoids common optical tweezers' calibration difficulties, system instabilities, as well as experimental and spatial restrictions.



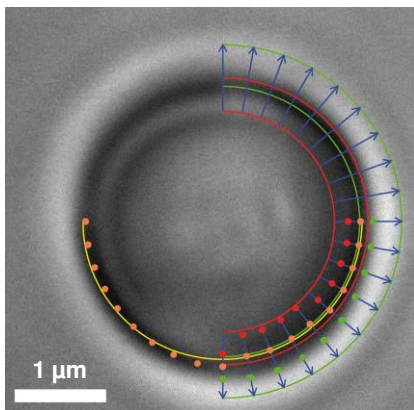
Left: The common principles of force measurement. Laser light of 1064 nm passing through the trapped particle (forward scattered light) is collected and projected onto a detector sensing the particle's deflection. The condenser in close proximity (ca. 1mm) to the trapping objective needs to be precisely adjusted. It limits the experimental space and is susceptible against drift and misalignment.

Right: In video-based particle detection the image of the trapped object - illuminated either by reflected or transmitted light - is directly projected onto a camera. This extremely robust setup allows high experimental freedom, where no detector alignment is required anymore, and an easy integration of fluorescence illumination. In addition, a great diversity of trapped particles can be video-analyzed and measured.

## General Aspects of PicoTweezers

- Quantitative 3D force measurements with 0.2 pN resolution (at 150 Hz sample rate)
- Very fast, easy and reliable force calibration
- No detector alignment or adjustment is required in the beginning or during experimentation
- Achievable trapping force of 300 pN with 1 W 1064 nm fiber laser
- Manipulation of trapped objects with nanometer precision
- Compact and ultrastable modular tweezers system
- Programmable LabView™ software interface
- Stand-alone system that can also be customized to any inverse microscope

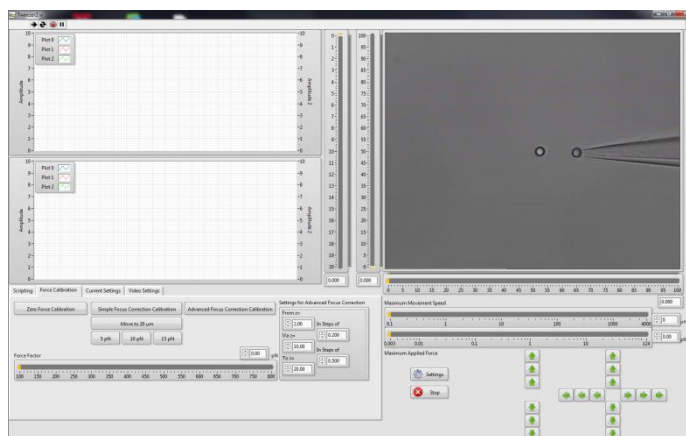
The entire video detection is integrated into the optical pathway between laser and optical trap. Since the diameter of the trapped object is permanently monitored, any particles of interest can be trapped and compared with previous ones on demand. Video detection is unsusceptible to disturbing particles that occasionally may be trapped together with the measured object. When trapping particles of interest close to interfaces (bottom or ceiling of a sample chamber, artificial or biological membranes, etc.), video analysis delivers an interference-free force signal.



Video frame of a trapped microbead with different overlaid detection lines.

## Video Detection and Analysis

The PicoTweezers system includes two cameras. One is imaging the surrounding area of the optical trap, and a second high-speed CMOS camera simultaneously surveys the magnified image of the trapped particle. The software analyzes both the position and size of the particle in real-time by edge detection and translate them into a force.



LabView™ based trapping, calibration and measurement software.

## Easy and Reliable Force Calibration

Force calibration in three dimensions is conducted by an automated routine moving the surrounding medium via the 3D piezo stage using Stokes' drag force law.