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Constructions and Applications of a Simple Optical Tweezers

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The combination of laser with optical microscope provides the exciting possibility of not only observing small objects but also trapping or even manipulating them. "A Single Beam Gradient Force Trap", also called "Optical Tweezers", uses a strongly focused laser beam to create a trapping zone, which is stable in all three dimensions. (Ashkin 1986) The axial stability of the trap is derived from the large axial gradient in light intensity near the focus. It gives rise to a gradient force (also called " negative radiation pressure"), capable of pulling a particle back into the trapping zone (the largest light intensity area), even against the direction of light propagation. (Block) In the aqua, optical tweezers had been demonstrated to trap dielectric particles with dimensions between 25 nm to 10 μm. (Ashkin et al. 1986) It has been used to manipulate small objects such as motile bacteria and sperm cells, to move eurkaryotic cells through the fluid, and to displace certain vesicles and organelles within larger cells. (Ashkin et al. 1987, Block et al. 1989) Recently, optical tweezers has been applied to study the DNA dynamics. (Perkins et al. 1994).

The schematic of our optical tweezers is shown in Fig. 1. In order to ensure enough gradient force, it is necessary to have a large gradient field around the trapping zone. This is accomplished by focusing the laser beam by a large N.A. objective. A trapping zone is formed near, but not at the focus. In addition, the same microscope is used to observe the trapping phenomena. The laser is an Argon laser operated at 514.5nm and the microscope is the Nikon Optiphot-2. The microscope has two auxiliary ports: the video output port, and the epi-fluorescence attachment port. For precisely adjustments and measurements, the video monitor is more helpful than the eyepieces. The laser beam is injected into the microscope through the epi-fluorescence attachment port. The microscope objective is a 100×ELWD Plan Achromat with N.A. 0.8.

Some modifications are made to the Nikon Optiphot-2 microscope. (Afzal 1989) The laser beam passes through a positive lens before entering the epi-fluorescence attachment port to insure the focus falling onto the object plane of objectives.

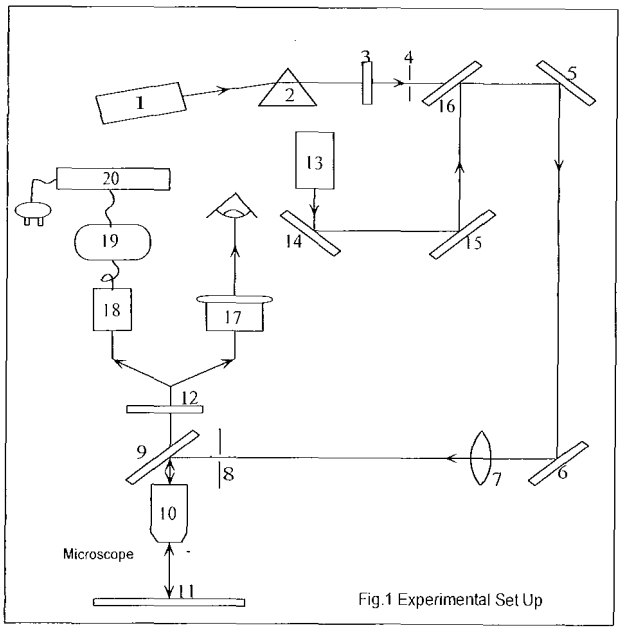

Fig. 1. The experimental set up contains two parts: Part (I) the optical tweezers and Part (II) the observation System.

Part (I)
1.Argon laser
2.Prism
3.ND filter
4.Iris
5.Reflection mirror
6.Reflection mirror
7.Positive lens
8.Epi-fluorescence attachment port
9.Dichroic mirror
10.Objective
11.Stage
12.Color filter
13.He-Ne laser
14.Reflection mirror
15.Reflection mirror
16.Slide glasses
Part (II)
17.Eyepieces
18.CCD camera
19.Monitor
20.Video recorder

This allows simultaneously observation of both objects and laser spot. In addition, the laser spot could be minimized by adjusting the position of this lens. To reflect the laser beams down to the objectives, a dichroic mirror is placed between the epi-fluorescence attachment port and the objectives. This dichroic mirror allows some portion of white light from microscope going through, preserving the original function of the microscope. A long-pass color filter is added to block the laser light from saturating the CCD camera. A low power He-Ne laser is collinear with the Argon laser to serve as the indicator for the laser focus.

Polystyrene latex is chosen to test the optical tweezers because of its spherical shape and uniform size. Trapping of 4μm polystyrene latex could be obtained using power as low as 1mW, and the trapped polystyrene latex can be pulled freely within the field of view. We have estimated the trapping force by the following method. By moving the stage of the microscope continuously, we observed that other objects moved rapidly with respect to the trapped particle. The velocity of this motion enabled us to estimate the trapping force because most of the trapping force is used to overcome the viscous force of water. The viscous force of water can be obtained from the Stock's law $f = 6\pi\eta Ur$, where η is the viscosity of water, U is the velocity of the trapped particle, and r is the radius of the particle. By putting proper numbers into the formula, the trapping force is estimated to be on the order of 0.1 to 0.01 μdynes. However, because of the difficulties in measuring the actual velocity, the trapping force should be larger than the above estimations. In addition, since a polystyrene latex with diameter 1.05 g/cm^3 will experience a gravity of 0.03 μdynes. Thus, the trapping force is hard to overcome the gravity. This helps to explain why the optical tweezers need to work in the aqua.

We had also used the optical tweezers to study Euglena. The Euglena, which belongs to the Mastigophora of the Protozoa, has the dimensions about 2×4 μm and one flagella. It can swim freely in the fluid. The optical tweezers can trap a Euglena with laser power about 100mW. In addition, the trapped Euglena could be pulled freely by the tweezers.

Other unconventional ways of constructing optical tweezers as well as the possibility of integrating the optical tweezers with scanning probe microscope and confocal optical microscope will also be discussed.

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