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Transport of microscopic objects using asymmetric transverse optical gradient force

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ABSTRACT We report an efficient technique based on an optical tweezers setup for optically controlled transport of microscopic objects. The technique makes use of an elliptically profiled trap beam that has an asymmetric intensity distribution about the center of its long axis. Microscopic objects pulled into the trap from the side having the larger intensity gradient become accelerated along the major axis of the focus and are ejected from the lower-stiffness end. The speed of transport is determined by the laser-beam power and the degree of asymmetry in the intensity profile. The approach could be used to simultaneously trap and transport hundreds of particles, varying in sizes from sub-micrometer to a few micrometers. Further, transport of red blood cells using this method is demonstrated.

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1 Introduction

Transport of microscopic objects plays a very important role in several biophysical processes like signaling, locomotion, and so on, that are crucial for functioning of a cell [1]. Therefore, the ability to effect controlled transport of intracellular objects in a cell cannot only help understand these fundamental processes but also provide a means to manipulate the functionality of living cells. Development of techniques for controlled transport of microscopic objects is also of considerable interest from the point of view of their use in microfluidic devices [2]. There have been several reports on the use of the axial light scattering force from a weakly focused or collimated laser beam for transport of particles along the direction of the beam [3–5]. If the scattering force dominates the axial gradient force, the particle is propelled along the direction of the beam. The dependence of the scattering force on size and refractive index has also been exploited for separation of particles in a flowing liquid by having the laser beam propagate in a direction opposite to the direction of liquid flow [6]. The particles become stationary (trapped) when the drag force exerted on the particles by the liquid flow balances the scattering force. However, this approach cannot be used for transport of particles in a plane transverse to the laser-beam propagation that is often desirable for manipulation and study of samples under full microscopic view. Transport of trapped objects, in a plane transverse to the laser-beam propagation, has been carried out either by scanning [7] the trapping beam or by keeping the trapping beam(s) fixed and moving the stage. A major limitation of this approach is that only a handful of objects can be manipulated at a time, limited by the maximum time that could be allowed before the trap beam visits an object to ensure that the object does not diffuse away from the desired direction. Short repetitive sequences of holographic trapping patterns have also been used [8] to transport microscopic objects through large arrays of optical tweezers. This method is also limited by the update rate of the spatial light modulator. In this report, we describe the use of a laser trap beam having an asymmetric intensity distribution about the center of its long axis for efficient controlled transportation and acceleration of microscopic objects in a plane transverse to the laser-beam direction. The approach could be used to transport simultaneously a large number of particles varying in sizes from sub-micrometer to a few micrometers. The approach could also be used to effect transport of actin monomers in neurons and thus either accelerate the growth rate of the existing cones or generate new growth cones.

2 Experimental details

In the conventional laser tweezers the intensity distribution of the trap beam is symmetric about the beam center (Fig. 1a). The gradient forces in such a trap beam lead to a symmetric potential well (Fig. 1b). However, by creating asymmetry in the intensity distribution of the trap beam (Fig. 1c), an asymmetric potential well can be created (Fig. 1d). Particles at the steep end of such a potential well will be pulled towards the potential minimum, get accelerated, and be ejected along the direction having the lower stiffness. The speed with which particles move will be determined by the depth and the asymmetry of the potential well, which can be controlled by the trap beam

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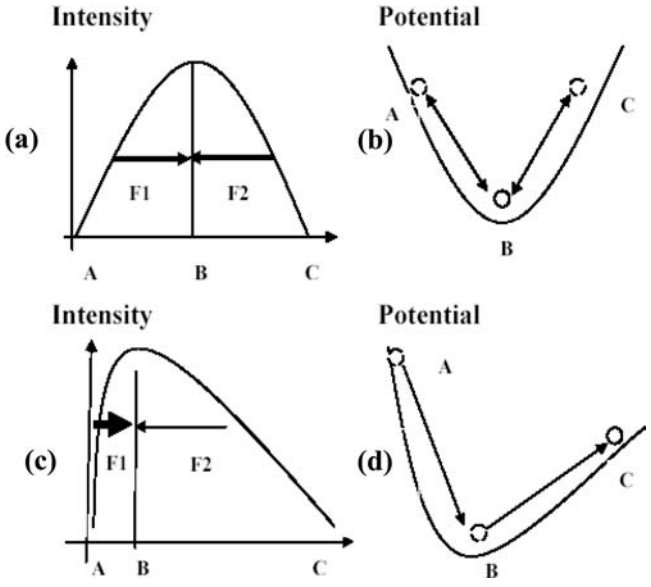

FIGURE 1 Optical transport with asymmetric trap beam. In a symmetric elliptical trap (**a**) the intensity gradient is symmetric about the center of the beam (B), leading to symmetric restoring forces F1 and F2. Therefore, a particle will be trapped at the center of the trap beam (**b**). In an asymmetric elliptical trap (**c**) the intensity gradient is steeper from (**a**) to (**b**) compared to that from (**c**) to (**b**), leading to asymmetric gradient forces F1 and F2. Therefore, particles at the steep end of the potential well will be pulled towards the potential minimum, become accelerated, and be ejected along the direction having the lower stiffness (**d**)

power and the degree of asymmetry in the intensity profile.

For realization of the above-discussed approach for transport of objects we used a line optical tweezers setup where a 1064-nm continuous-wave Nd:YAG laser operating in TEM_{00} mode served as the trapping beam. The laser beam was expanded using a beam expander, passed through beam-steering and aligning mirrors, and coupled to a $\times 100$ microscope objective (Zeiss Plan Neofluor phase objective $\times 100/1.3$, oil-immersion type) through a combination of cylindrical and spherical lenses to focus it to an elliptical spot in the specimen plane. The focal length of the cylindrical lens was changed to vary the dimensions of the elliptical beam profile at the object plane. In our setup [9], use of a 200-mm-focal length cylindrical lens led to a focal spot size at the object plane of $\sim 1\,\mu m \times 10\,\mu m$ and with a 50-mm-focal length cylindrical lens the focal spot size at the object plane was $\sim 1\,\mu m \times 40\,\mu m$. For all the experiments reported here we coupled up to ~ 200-mW power to the microscope objective. Intensity asymmetry along the major axis of the elliptical focus was created by controlling the angle that the laser beam makes with the optical axis of the cylindrical lens(es). A stepper motor was used to rotate the cylindrical lens mount and thus the direction of the major axis of the elliptical focal spot. A dichroic beam splitter was used to reflect the transmitted visible light to a charge-coupled device (CCD) that was used to visualize the trapping and transport of the microscopic objects. To prevent the back-scattered laser light reaching the CCD detector, an IR cut-off filter was used. The motion of the trapped object was recorded on a video cassette using a video cassette recorder. These images were digitized using a frame grabber and a computer. The translation speed, acceleration, and so on, were measured by analyzing the position of the moving object(s) in successive frames. For mapping the intensity profile of the elliptical focus, back-scattered laser light of a probe beam (He-Ne laser, pre-aligned with the Nd:YAG laser beam) from the particles was also recorded.

It is pertinent to emphasize here that in our experiments an asymmetric intensity distribution was created only in the direction of the major axis of the elliptical trap beam. In the other two directions, i.e. the direction of propagation of the laser beam and the minor axis of the elliptical trap, the intensity distribution was symmetric, resulting in symmetric gradient forces which led to trapping of the objects, as in conventional optical tweezers.

3 Results and discussion

In Fig. 2 we show the digitized time-lapse video images of optical transport of a silica particle of size $\sim 5\,\mu m$ when the asymmetric trap

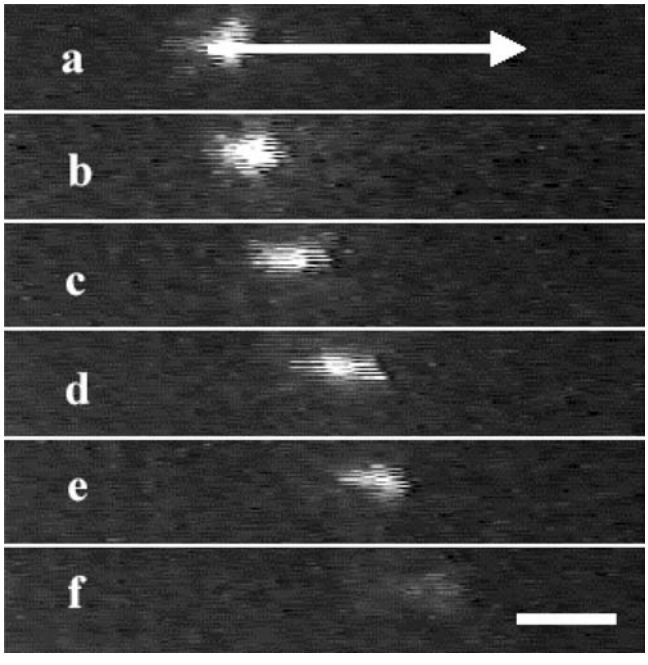

FIGURE 2 Digitized time-lapse video images of the transport of a silica particle of size $\sim 5\,\mu m$. Because of the asymmetry of the trap beam intensity profile the intensity of the back-scattered light increases from (**a**) to (**b**) and then reduces from (**b**) to (**f**). The time lapse in consecutive frames (**a**)–(**f**) was 80 ms. All images are in same magnification. Scale bar: 10 μm

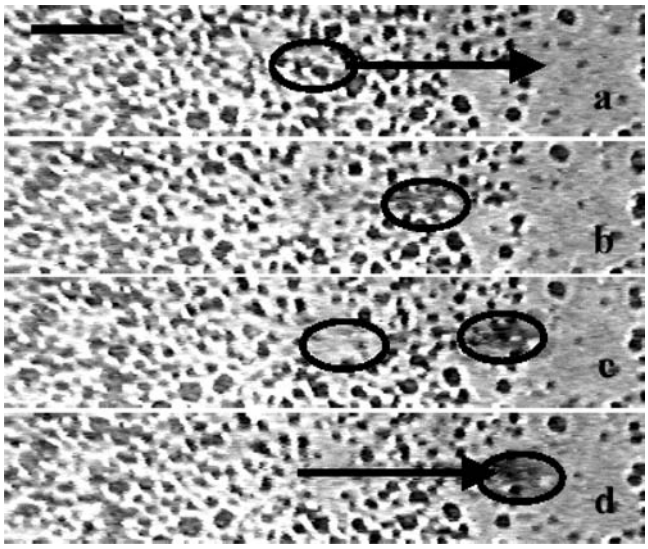

FIGURE 3 Bright-field digitized time-lapse video images of transport of a mixture of silica particles of sizes varying from 1 to 5 μm. The time lapse in consecutive frames (**a**)–(**d**) was 200 ms. All images are in same magnification. Scale bar: 20 μm

beam (1 μm × 40 μm) was switched on. The silica particle placed on the cover slip was brought close to the steeper intensity gradient end of the elliptical focal spot whose location is indicated in Fig. 2a by an arrow. The direction of the arrow points in the direction of transport and the length of the arrow corresponds to the length of the major axis of the asymmetric trap beam. Since the particle is visualized by its back scattering of the He-Ne probe beam, the back-scattered intensity in the time-lapse video images of the particle represents the trap beam intensity profile. The back-scattered intensity is seen to increase for a short distance (about 2 μm from the left-hand side) and then decreases slowly (over 35 μm), which is consistent with the intensity distribution of the trap beam.

In Fig. 3, we show the use of the asymmetric trap beam for transportation of a collection of silica particles (Sigma Chemicals) of sizes varying from 1 to 5 μm. The distribution of particles before the trap beam is switched on is shown in Fig. 3a. Here also the position and length of the arrow correspond to the location and length of the major axis of the trapping beam and the direction of the arrow points in the direction of transport. In Fig. 3a, very few particles are seen at the tip of the arrow. With the transporting beam switched on, the particles are transported towards the shallower intensity gradient end of the elliptical focal spot and accumulate there (Fig. 3b to d). With 200 mW of beam power at the specimen stage we could simultaneously transport a few hundred particles at a speed of ∼ 20 μm/s along the 40-μm-long major axis of the elliptical profile. Transport of a collection of polystyrene nanoparticles of sizes ∼ 200 nm was also achieved with the technique. However, in this case, because of the small size of the particles the back-scattered intensity was very weak.

The technique allows control of both the direction of transport as well as the speed of transport. The direction of transport can be varied by rotating the cylindrical lens to fix the direction of the major axis of the elliptical focus at the desired angle in the transverse plane [9]. The speed of transport can be controlled by the trap beam power and the degree of asymmetry in the intensity profile. In Fig. 4, we show the change effected in the transport of a single red blood cell by controlling the trap beam power. The speed of transport could be increased from ∼ 35 μm/s at 38-mW trap beam power to ∼ 100 μm/s at 175 mW.

It is important to note that the asymmetric optical gradient forces, which determine the magnitudes of acceleration and velocity of the particles, depend on the optical and geometrical properties of the particles. The technique can therefore be used for in situ sorting of different particles based on differences in their physical properties (either size or optical properties). We should emphasize that the present approach can be used for all-optical microfluidic processing [10] and does not require any fluid flow, as is the case in the recent reports of optical fractionation [11, 12] or optical chromatography [6] that exploit the difference in interactions of particles flowing in a fluid with optical tweezers for separating the particles.

We have also investigated the use of this approach for transport of intracellular objects. For this purpose experiments were carried out on neuronal cells. This choice was made for two reasons. First, the neuronal growth is believed to involve transport of actin monomers in the direction of the growth cone [13]. Secondly, the use of a transverse gradient force directed towards the center of a laser trap beam, achieved by positioning a defocused laser-beam spot ahead of the growth cones, has already

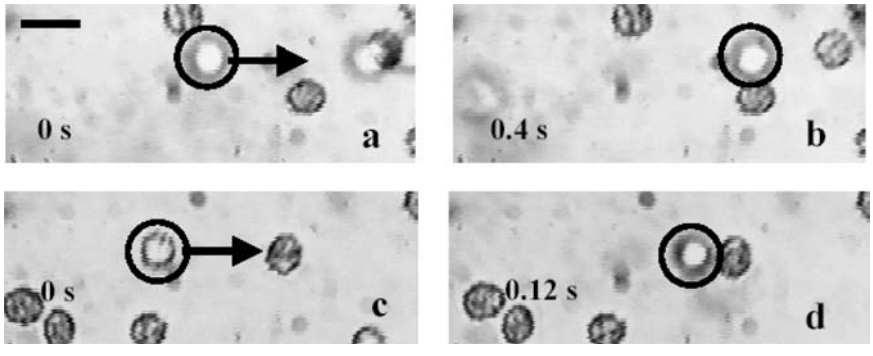

FIGURE 4 Bright-field digitized time-lapse video images of transport of red blood cell (shown *encircled* for clarity) at two trap beam powers. The direction of the *arrow* points in the direction of transport and the length and the position of the arrow correspond to the length and the position of the major axis of the asymmetric trap beam. For (**a**) and (**b**), the trap beam power was 38 mW and, for (**c**) and (**d**), the trap beam power was 175 mW. All images are in same magnification. Scale bar: 10 μm

been shown [14] to accelerate the growth of a neuronal growth cone. We were able to achieve the same result, more efficiently, by use of an asymmetric trap beam to effect transport in the direction of the growth cone. In addition to the acceleration of the growth rate of the existing cones, the approach could also be used to generate new growth cones. Details of these results will be presented elsewhere.

We should also note that it is possible to have several interactive and real-time user-configured asymmetric intensity profiles which can be driven independently to simultaneously transport, mix, or even sort microscopic objects. Such devices by themselves or in combination with other microfluidic devices may find a wide range of applications in cell and molecular biology, colloidal science, and optically driven micromachines.

4 Conclusions

To conclude, we have developed a laser-assisted technique that is simple to implement and allows controlled transportation of microscopic objects along any desired direction in the plane transverse to the direction of propagation of the laser beam. Efficient transport of an ensemble of particles with dimensions ranging from sub-micron to a few microns has been achieved in the transverse plane. The approach could also be used to enhance the growth rate of existing neuronal cones as well as to generate new growth cones.

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REFERENCES

1 T.M. Svitkina, A.B. Verkhovsky, K.M. McQuade, G.G. Borisy, J. Cell Biol. **139**, 397 (1997)
2 A. Terray, J. Oakey, D.W.M. Marr, Science **296**, 1841 (2002)
3 A. Ashkin, Phys. Rev. Lett. **24**, 156 (1970)
4 T.N. Buican, M.J. Smyth, H.A. Crissman, G.C. Salzman, C.C. Stewart, J.C. Martin, Appl. Opt. **26**, 5311 (1987)
5 M. Uchida, M. Sato Maeda, H. Tashiro, Curr. Biol. **5**, 380 (1995)
6 S.J. Hart, A.V. Terray, Appl. Phys. Lett. **83**, 5316 (2004)
7 K. Visscher, G.J. Brakenhoff, J.J. Krol, Cytometry **14**, 105 (1993)
8 B.A. Koss, D. Grier, Appl. Phys. Lett. **82**, 3985 (2003)
9 R. Dasgupta, S.K. Mohanty, P.K. Gupta, Biotechnol. Lett. **25**, 1625 (2003); see also Opt. Photon. News **14**, 16 (2003)
10 J. Gluckstad, Nat. Mater. **3**, 9 (2004)
11 P. Korda, M. Taylor, D. Grier, Phys. Rev. Lett. **89**, 128 301 (2002)
12 M. MacDonald, G. Spalding, K. Dholakia, Nature **426**, 421 (2003)
13 B.K. Mueller, Annu. Rev. Neurosci. **22**, 351 (1999)
14 A. Ehrlicher, T. Betz, B. Stuhrmann, D. Koch, V. Milner, M.G. Raizen, J. Kas, Proc. Natl. Acad. Sci. USA **99**, 16 024 (2002)