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Manipulation of microparticles and red blood cells using optoelectronic tweezers

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Abstract. We report the development of an optoelectronic tweezers set-up which works by light-induced dielectrophoresis mechanism to manipulate microparticles. We used thermal evaporation technique for coating the organic polymer, titanium oxide phthalocyanine (TiOPc), as a photoconductive layer on ITO-coated glass slide. Compare to the conventional optical tweezers, the technique requires optical power in μW range and provides a manipulation area of a few mm^2. The set-up was used to manipulate the polystyrene microspheres and red blood cells (RBCs). The RBCs could be attracted or repelled by varying the frequency of the applied AC bias.

Keywords. Optoelectronic tweezers; optically-induced dielectrophoresis; AC dielectrophoresis.

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Optoelectronic tweezers (OET) is an optical manipulation technique that utilizes light-induced dielectrophoretic forces for micromanipulation and was developed in 2005 [1]. The technique requires an optical power in μW range and can manipulate thousands of cells/colloidal particles in an area of a few mm^2. In OET, the chip consists of an indium tin oxide (ITO)-coated glass substrate (top) and an ITO-coated glass substrate (bottom) with a photoconductive layer separated by spacers. An AC bias is applied between the electrodes and an intensity pattern is projected on the photoconductive layer, which generates virtual electrodes resulting in a non-uniform electric field between the electrodes. This non-uniform electric field induces dielectrophoretic forces on the particles in the OET chip.

In the development of the OET chip, the coating of a suitable photoconductive material on a glass substrate having an optically transparent conductive thin film layer (usually indium tin oxide, ITO) is a major hurdle. The material should have good photoconductivity, good adherence to the ITO film for coating, stable optical properties for at least a few days and be fabricated as a film by an easy method. In the first report, Ohta *et al*

[1] used an amorphous silicon (a:Si) layer on an ITO glass substrate as a photoconductive layer fabricated by plasma-enhanced chemical vapour deposition (PECVD) process, which is a relatively costly, high-temperature and complex procedure. For better adherence, a:Si also requires a thin layer of aluminum/molybdenum and a passive coating on the top to protect it from water. Therefore, several other approaches were used to replace the a:Si layer with an appropriate photoconductive material to simplify the fabrication of the OET chips. Recently, Wang *et al* [2] used three thin films PEDOT:PSS/P3HT:PCBM/LiF in sequence on an ITO-coated glass substrate to use polymer P3HT:PCBM as a photoconductive layer. Yang *et al* [3] reported a simpler approach to use a single layer of an organic polymer titanium oxide phthalocyanine (TiOPc) as a photoconductive material fabricated by spin coating. Though spin coating of TiOPc on an ITO glass substrate is a relatively easier process, for a good quality film a highly homogeneous solution of TiOPc is required, which is a tedious job as TiOPc is insoluble in inorganic solvents and has very low solubility in organic solvents [4]. A technique that can use TiOPc in a powder form will be more suitable for coating. We report the development of an OET device that uses a photoconductive TiOPc layer on an ITO-coated glass substrate fabricated by thermal evaporation technique. The thermally deposited photoconductive layer has very good adherence to the ITO film, obviating the need for an intermediate layer of any material between the ITO film and the photoconductive layer to improve the adherence of the photoconductive material. The set-up was used for manipulating polystyrene microspheres and red blood cells (RBCs). We observed that RBCs were getting attracted to the illumination region at ∼200 kHz frequencies or higher and repelled at ∼80 kHz or lower frequencies of the applied AC bias.

A schematic of the OET chip is shown in figure 1. The liquid containing the sample is sandwiched between an ITO-coated glass slide (top) and another ITO-coated glass slide (bottom) containing a ∼200 nm TiOPc layer fabricated using thermal evaporation (∼10^{-5} torr) technique. We observed that the TiOPc layer has good adherence to the ITO

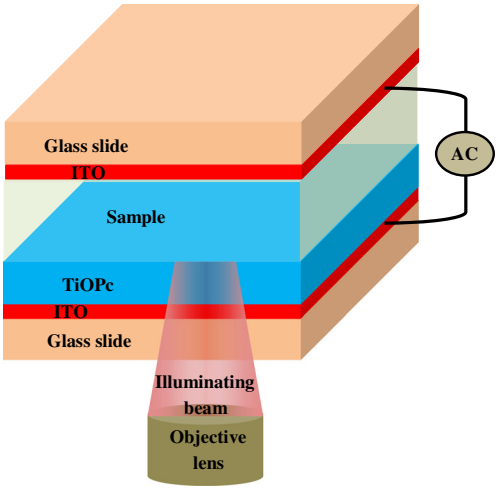

Figure 1. A schematic of the OET chip.

film and does not require coating of any intermediate layer between the ITO film and the photoconductive material for adherence. The top glass slide is flipped over and put on the bottom slide with a $\sim$100 μm spacer.

The OET chip was used to perform experiments using a set-up shown in figure 2. A 664 nm diode laser (ILX Lightwave) with an output power of 150 mW was used to illuminate the liquid crystal-based spatial light modulator (SLM) (LCR 2500, Holoeye) for generating the dynamically reconfigurable multiple optical traps. An aspheric lens L1 was used to collimate the laser output, which was further spatially filtered using a pair of lenses (L2, L3) and a pin hole aperture (A). The same lens pair was also used to expand the laser beam suitably before falling on the SLM. The SLM was placed in the conjugate plane to the input pupil of a 10X objective lens using a lens pair (L4, L5) of 500 mm focal length each. A fibre-optic illumination source (Dolan Jenner) was used for bright field illumination. The images were collected using 25X objective lens and were focussed on to a monochrome CCD (Oscar) using a lens (L6). A short pass filter (F) was used to suppress the laser light to the CCD. The AC biasing on the ITO electrodes was applied by a function generator (Agilent 3220). For experiments $\sim$15 μm polystyrene spheres (Polysciences Inc.) were suspended in deionized water with 1% fetal bovine serum (FBS), which prevents the adhesion of the polystyrene spheres on the photoconductive surface. RBCs were suspended in an isotonic solution consisting of 8.5% sucrose, 0.3% dextrose in deionized water making conductivity of the solution as $\sim$10 mS/m using KCl.

Optoelectronic tweezers traps microparticles by dielectrophoresis (DEP) force, which is given by (for spherical particle) [5]

$$F_{\text{dep}} = 2\pi r^3 \varepsilon_m \text{Re}\left[K^*(\omega)\right] \nabla E^2, \tag{1}$$

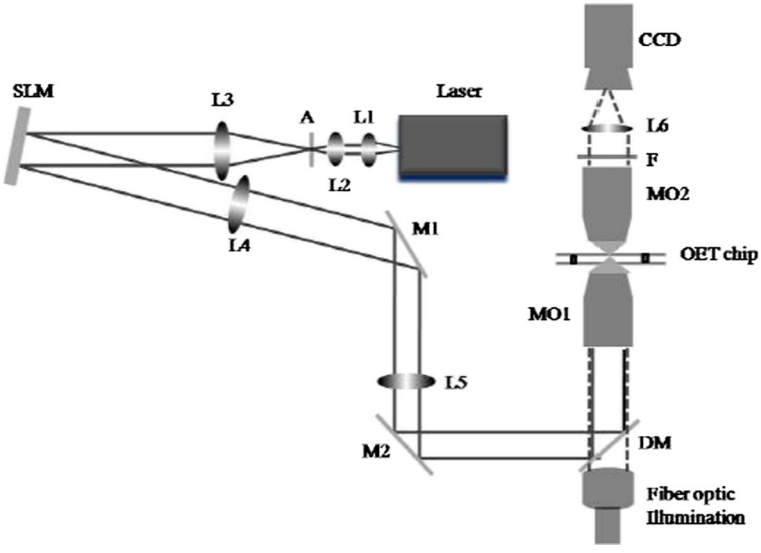

Figure 2. A schematic of the experimental set-up.

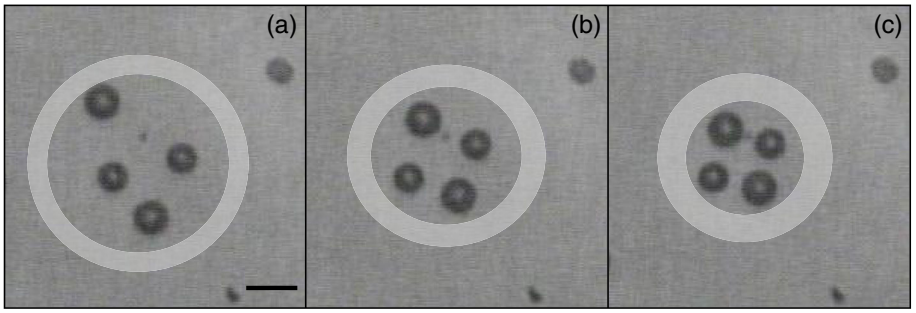

Figure 3. Sequential images showing the particle concentration using varying mode of LG beam. The rings showing the schematic LG beam intensity pattern for (**a**) LG_{25}, (**b**) LG_{18} and (**c**) LG_{10}. Scale bar: 30 μm.

where

$$K^*(\omega) = \frac{\varepsilon_p^* - \varepsilon_m^*}{\varepsilon_p^* - 2\varepsilon_m^*}, \qquad \varepsilon_p^* = \varepsilon_p - j\frac{\sigma_p}{\omega},$$

and

$$\varepsilon_m^* = \varepsilon_m - j\frac{\sigma_m}{\omega}, \tag{2}$$

where r is the radius of the particle, ε_m is the permittivity of the media, E is the electric field and $\mathrm{Re}[K(\omega)]$ is the real part of the Clausius–Mossotti factor, σ_p and σ_m are the conductivities of the medium and the particle respectively and ω is the angular frequency of the applied electric field. If the particle is more polarizable than the medium, that is, $\mathrm{Re}[K^*(\omega)] > 0$, it is called positive DEP and the particle is attracted towards the higher electric field region. If the particle is less polarizable, $\mathrm{Re}[K^*(\omega)] < 0$, it is called negative DEP as the direction of the DEP force is towards lower electric field region.

The applied AC bias was 10 V_{pp}, 100 kHz and the total power at the sample plane was $\sim$8 μW. The polystyrene microspheres will always have negative DEP, and so to trap them an annular intensity pattern (Laguerre Gaussian (LG) beam) was used and the particles were translated up to a speed of $\sim$8 μm/s by translating the LG pattern (LG_{10}). Further, LG beams of variable mode orders were used for concentrating the particles, as shown in figure 3. As the mode order of LG beam was changed from LG_{25} to LG_{10} in a

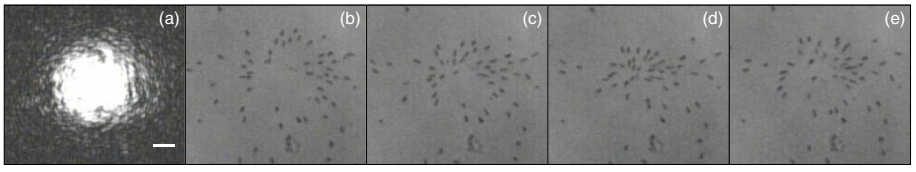

Figure 4. Manipulation of RBCs (**a**) illuminated intensity pattern, (**b**)–(**d**) RBCs getting attracted towards the illuminated region and (**e**) RBCs getting repelled from the illuminated region. Scale bar: 25 μm.

sequential way, the microspheres being repelled by the intense region was concentrated in the dark central region of the LG beam intensity profile, as shown in figures 3a–3c.

When a Gaussian intensity pattern, as shown in figure 4a, was applied to manipulate the RBCs at 200 kHz, 10 V_{pp}, the cells were attracted towards the illuminated region, as shown in figures 4b–4d. During the process, when the frequency of the applied bias was changed to 80 kHz, the cells started repelling slowly and at 40 kHz, cells repelled very fast from the illuminated region, as shown in figure 4e. By changing the frequency, the cells repelled as the Clausius–Mossotti factor changed sign from positive to negative, thus inducing a negative DEP force [6].

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