Mutual influence of time-shared optical traps studied by means of Video Holographic Microscopy

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Holographic microscopy and calculations in three dimensions are used to explore how properties of an optical trap, such as stiffness and trapping position, are influenced by other traps in its vicinity. The influence of individual traps is not limited to their diffraction-limited spot in the trapping plane, and creating a true 1D line trap is not straightforward. Here, we use a Mie–Debye representation, including the effect of spherical aberrations, to calculate the force fields. We compare the calculations with experiments on a single particle in a linear array of three traps. We vary the spacing between the traps and measure the particle's trajectory using video holographic microscopy. Thus we can study the effect of nearby traps on the trapping position and trapping stiffness in 3D. We find almost quantitative agreement between theory and experiments which allows us to predict the effect of arrays of optical traps. ©2009 Optical society of America
OCIS codes: (090.1995) Digital holography; (110.0180) Microscopy; (180.3170) Three-dimensional microscopy; (230.1040) Acousto-optical devices; (290.4020) Mie theory; (350.4855) Optical tweezers or optical manipulation.

1. Introduction

With optical tweezers, a dielectric particle with a refractive index larger than that of the medium can be trapped [1]. Using acousto-optical deflectors (AODs) one can create arbitrary arrays of point traps [2]. These can be used to trap e.g. nanorods [3], create local defects in photonic crystals and in combination with confocal microscopy [4] they can be used to make patterns to study colloidal epitaxy [5]. Here we combine time-share traps with holographic microscopy.

A particle in an array of time-shared traps experiences a time averaged potential landscape. If the time-sharing is fast enough the particle will remain trapped [2,5]; the particles will not diffuse away by Brownian motion. Time-sharing excludes the occurrence of interference between light of nearby traps. This does not mean these traps do not effect the trapping position and stiffness of each other. Due to the large opening angle of the high-numerical aperture (NA) objectives used, the effect of point traps is not limited to the diffraction limited spot. This can be used by making a linear array of traps to form a line trap, for instance to measure interaction forces [1a]. The vicinity of neighboring traps then makes the stiffness diminish in one direction. However, the neighboring traps also influence the trapping depth and often also reduce the stiffness in the other directions. In this work, calculations of these effects in a (simple) line trap configuration are presented, together with experiments on the same array of traps.

2. Theory

The calculations of the forces exerted by single traps use the Mie–Debye representation first given by Maia Neto and Nussenzveig for the axial direction [7], expanded to 3D by Mazolli et al. [8], and complemented by Viana et al.[9] to include spherical aberrations. The parameters used to describe the traps are the beam opening angle θ and γ, the ratio of the objective focal length to the beam waist ω_0. We used $\theta = 64.245°$, $\gamma = 1.21$, and laser wavelength $\lambda = 1064$ nm. The spherical aberrations result from the refractive index contrast between the cover slip and the solvent and depend on the distance from the geometric focus to the glass surface; here a distance of 12 μm was used.

The use of time-sharing to create multiple traps implies that there is never more than one trap and that there is no interference between the traps in the array. Therefore, we can add the forces of the individual traps [2, 3, 6]. To calculate the force-field around a line-trap we calculate the forces exerted by one trap on a grid around it. We assume spherically polarized light reducing the problem to 2D as a result of the cylindrical symmetry. This reduces calculating the total force around the array to looking up the distances to all traps and adding the forces they exert.

The same scattering behaviour that enables us to exert forces on the particles [10] is used to track the position of the particles in time. The scattered light interferes with the unaffected part of the illuminating plane waves to form a fringe pattern, a hologram. This hologram is magnified using the same high-NA objective we use for optical trapping and recorded using a high-speed CCD camera. Finding the particles' positions in 3D is then reduced to solving an inverse problem [11]. Because we have a quantitative model for the scattering [10] this yields a resolution of less than 1 nm in lateral directions and less than 10 nm in the axial direction [12]. This technique is known as video holographic microscopy and measures the particles' coordinates in 3D from a single 2D image. These can be recorded at video rate or even faster. The resulting 3D trajectories can be used to extract the stiffness of the optical trap by comparison to the proper Langevin equation [13].

3. Experimental details

To allow for video holographic microscopy in our setup [4] (Fig. 1a), the condenser of an inverted microscope (Leica DM IRB) is replaced by a 532 nm laser. An infrared laser beam (Spectra Physics Millennia, 1064 nm, 10W cw) is split at a polarizing beam splitter, while the rotation of the wave plate determines the power sent to the objective. Just before the objective (Leica 100x 1.4 NA oil immersion), a pair of lenses (both f = 80mm) forms a telescope to provide for manual displacement of the laser focus; for holographic microscopy the particles need to be trapped above the imaging focal plane. To enable time-sharing the laser beam passes a pair of AODs (IntraAction DTD-276HB6), positioned at a plane conjugate to the back focal plane of the objectives. The AODs are powered by direct digital synthesizers (Novatech DDS 8 m) controlled by a LabVIEW (National Instruments) program. The beam is expanded in two steps: before the AODs by a 6× beam expander and after the AODs ~2× by two lenses (f = 120mm and f = 250 mm, respectively).

The sample cell, made of two coverslips (Menzel No. 1), sealed with candle wax, was filled with a dilute dispersion of SiO_2 (n = 1.45, 0.7 μm radius) beads in ethanol and was positioned 12 μm below the focus of the trap.

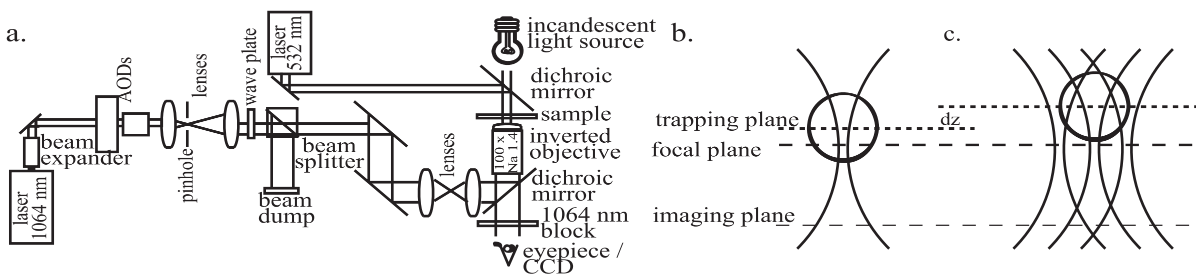

Figure 1. Schematic view of (a) the optical tweezers setup with adaptations for video holographic microscopy, (b) a particle trapped in an individual single-beam trap well above the imaging plane, (c) the same particle trapped in the middle one of three identical traps, some of the light of the neighbouring traps goes through the particle.

The experiment was conducted by trapping a particle in the middle one of the three traps. Then, while the hologram of the particle was recorded at 200 fps by the CCD camera (Mikrotron MC1362), the two outer traps were moved stepwise inward until all three traps overlapped. Simultaneously, a second particle was trapped well away (~ 8 μm) from the particle in the line array. This trap was moved simultaneously (in a square wave fashion to pinpoint the moments of stepping) parallel to the line at the same separation ***d*** from the final position as the outer traps of the line, allowing measurement of the exact separation.

4. Results

From the raw data in Fig. 2a can be seen that the particle remained trapped in the middle trap, but for separations around 1 μm the particle became trapped in a minimum somewhere in between two traps. However, the particle moved deeper in the sample for these separations. This is predicted by the calculations and is shown in Fig. 2b, where the average trapping position and the calculated position are plotted against the spacing. At these trap separations one expects the strongest effects as these are comparable to the particle diameter. In the axial direction (***z***) the effects are particularly large; the particle experiences some of the scattering force of two traps. Also the lateral displacement of the trapping position is in accordance with the calculations.

In Fig. 3c the measured trapping stiffnesses in the three directions [13] are plotted together with the calculation results. The stiffnesses in all directions decreased with a factor of three as the spacing between the three traps is increased from zero to a large distance, such that the power is divided over three traps. This can also be seen from the slightly less noise at these separations in the curves in Fig. 2a. For distances of about 0.3 μm there was a maximum in the trapping stiffness in the in-plane directions. This can be attributed to the fact that the effective focus at this spacing is more similar to the particle size. In fact our 1.4 μm diameter particles are larger than the optimal size for stiff trapping; see e.g. [7-9]. For a spacing of circa 1 μm there is quite a deep minimum in the stiffness in the direction of the line, ***y***. Thus for this spacing the line trap is working. However, this is also the distance at which the particle gets trapped the furthest in **z** from the normal trapping plane; the line is not straight. Since the particle was confined to a single position, although only for a short time interval, there are still barriers on the line preventing the particle to freely diffuse over the line. In this sense the line is also not 'straight'.

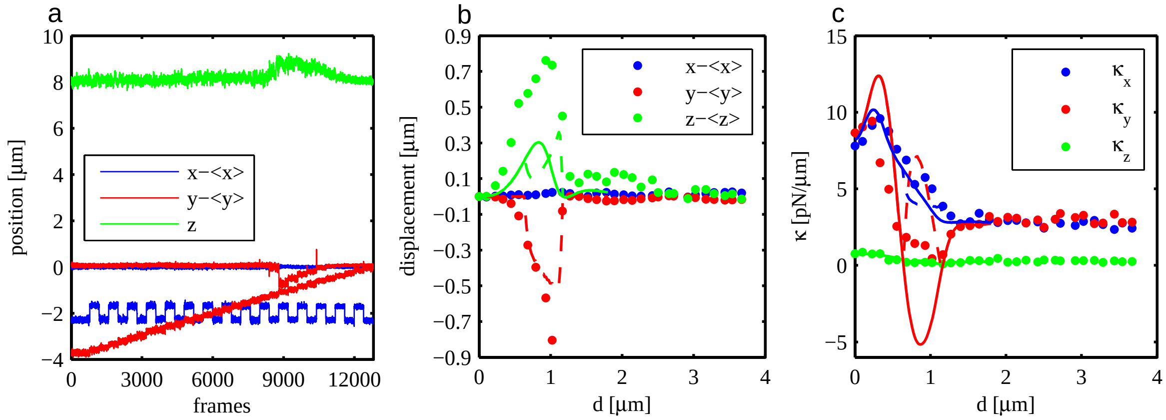

Figure 2. Graphs of (a) the raw particle trajectories, (b) the trapping position of the particle in the line trap as function of the trap spacing and (c) the stiffness κ in the x, y, and z directions. The power in the calculations of the stiffness were normalised to the value at zero spacing for the lateral directions and the axial direction separately. Symbols represent the measurements, full lines are the calculated values for the central trap of the line and dashed lines are the calculated values for the most probable trapping position.

To be able to perform holographic microscopy, the particles had to be trapped above the imaging plane of the microscope. This was accomplished using a slightly divergent laser beam to form the traps. We assume that this 'misalignment' is the main cause of the remaining differences between the experiments and the calculations. This is under current investigation.

5. Conclusions

Multiple time-shared optical point traps were modelled using the methods described by Viana et al. [9] and these calculations were found to be in agreement with experiments where the trap performance was monitored using video holographic microscopy [12]. The model can be used to predict the potential landscape formed by arrays of time-shared point traps. The results presented here for three traps illustrate the decrease in trap stiffness and increase in trapping depth for particles in an optical line trap.

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