

# Measuring and adjusting the trapping position in optical tweezers

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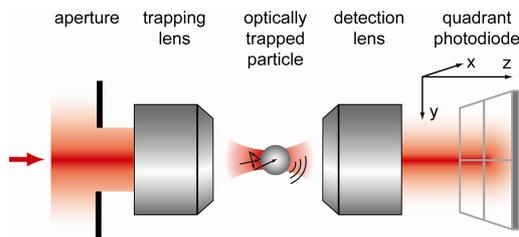
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We show how the trapping position of an optically trapped 1- $\mu\text{m}$  polystyrene bead is measured, and that it can be shifted by changing the relative radiation pressure exerted on the bead. The trapping position of the bead relative to the focus is measured using defocusing microscopy and 2-photon microscopy. Our observations are explained by the two component approach for optical forces.

## 1 Introduction

Optical tweezers have been widely used to trap and manipulate microscopic particles for about two decades [1]. Back-focal plane interferometry with a quadrant photodiode (QPD) enables 3D tracking of a trapped bead (Fig. 1) with nm precision at high sampling rates (up to one MHz) [2].



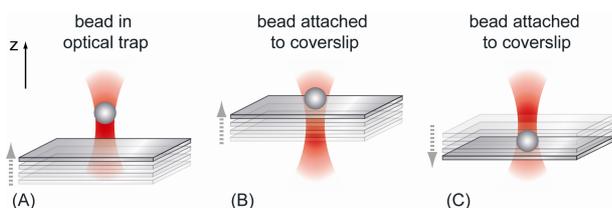
**Fig. 1** With a quadrant photodiode (QPD), an optically trapped bead is tracked in 3D.

As the 3D position signals from the QPD are only unique and linear in a restricted region around the bead's trapping position, it is crucial to know the position signals and the trapping position relative to these signals. As the bead is trapped on the optical axis, it is sufficient to measure the axial trapping position of the bead.

## 2 Trapping position relative to QPD signal

### 2.1 Measurement principle

The trapping position of a 1- $\mu\text{m}$  (diameter 1.03  $\mu\text{m}$ ) polystyrene bead trapped with an infrared laser ( $\lambda = 1064 \text{ nm}$ ) [2] was measured as described in [1] and [3] and shown in Fig. 2.

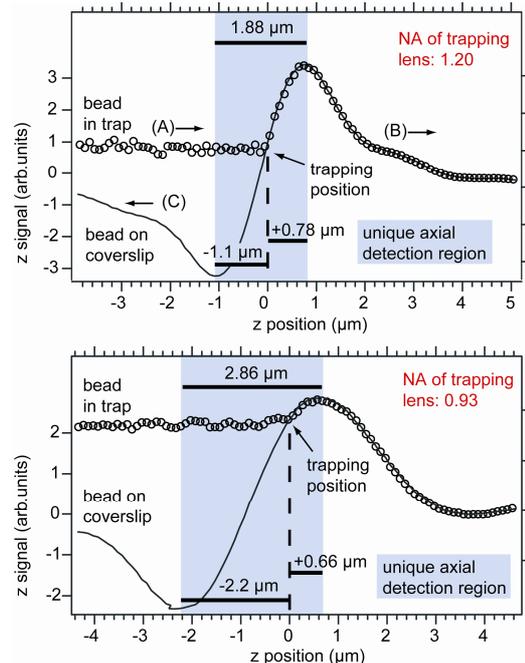


**Fig. 2** Determination of the bead's trapping position relative to the detector signal.

The bead is trapped above the coverslip (A). The coverslip is moved upwards and displaces the bead that attaches to the coverslip first in positive axial direction (B) and then in negative direction (C) while the QPD z-signal is recorded.

### 2.2 Results

The results of this measurement of the trapping position  $z_t$  relative to the detector z-signal are displayed in Fig. 3 for a high (1.20) and a lower (0.93) numerical aperture (NA) of the trapping lens.



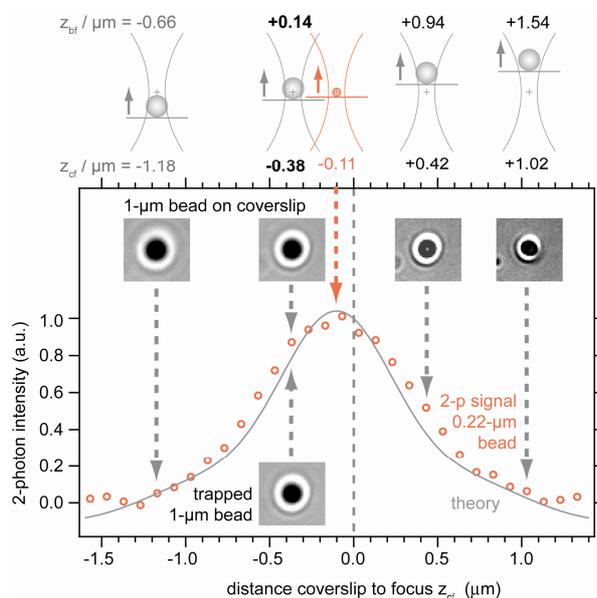
**Fig. 3** Measurement of the trapping position  $z_t$  relative to the QPD detector signal for the z position.

It can be seen that  $z_t$  is shifted along the optical axis in the direction of beam propagation if the trapping lens NA is decreased from 1.20 to 0.93. As the extent of the point spread function (PSF) increases with decreasing NA, the unique axial detector range (Fig. 3) and the unique lateral detector range (data not shown) also increase. A table summarizing these results for NAs ranging from 0.30 to 1.20 can be found in reference [3]. At

laser powers on the order of 10 mW in the focal plane, stable trapping of a 1- $\mu\text{m}$  bead is possible for trapping NAs ranging from 1.20 down to 0.74.

### 3 Trapping position relative to focus

We used defocusing microscopy and 2-photon microscopy as shown in Fig. 4 in order to determine the trapping position  $z_t$  relative to the (geometric) focus for a trapping NA of 1.20. The defocused brightfield image of a trapped 1- $\mu\text{m}$  latex bead was compared to a sequence of images of the bead fixed on a coverslip and moved in the axial direction through the focal region (inset pictures). By this method,  $z_t$  was determined relative to the coverslip position. The integrated fluorescence emission after 2-photon excitation at  $\lambda_{2p} = 532 \text{ nm}$  of a fluorescent 0.22- $\mu\text{m}$  bead fixed on the coverslip was measured as a function of the axial position of the coverslip (red circles). The gray curve shows the theoretical 2-photon signal of a point like object [4]. The 2-photon signal is maximal if the center of the 0.22- $\mu\text{m}$  bead is at the focus of the laser beam. This measurement determines the distance of the coverslip to the focus  $z_{cf}$ . At the position where the 1- $\mu\text{m}$  bead on the coverslip is at the trapping position, the coverslip is 0.38  $\mu\text{m}$  in front of the focus ( $z_{cf} = -0.38 \mu\text{m}$ ). As the 1- $\mu\text{m}$  bead has a radius of  $r = 0.52 \mu\text{m}$ , the distance of the bead center to the focus  $z_{bf}$  is therefore  $z_{bf} = z_{cf} + 0.52 \mu\text{m} = 0.14 \mu\text{m}$ : The 1- $\mu\text{m}$  bead is trapped 140 nm behind the focus.

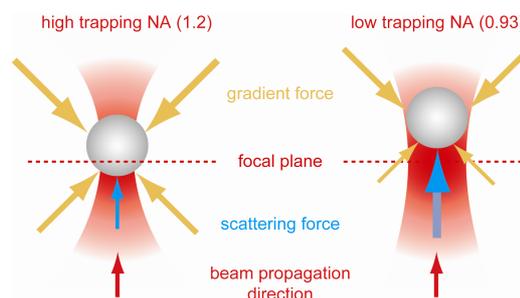


**Fig. 4** A 1- $\mu\text{m}$  bead is trapped 140 nm behind the focus for a trapping NA of 1.20. This was measured using defocusing microscopy and 2-photon microscopy.

### 4 Theoretical explanation

We found that a 1- $\mu\text{m}$  latex bead is trapped 140 nm behind the focus for trapping lens NA of 1.20 and that the trapping position  $z_t$  is shifted

along the optical axis for a lower NA. As shown in Fig. 5, both observations can be explained by using the two component approach for optical forces, where the total force acting on a small particle is written as the sum of the gradient and the scattering forces [2]. The gradient force is proportional to the gradient of the focal intensity and pulls a bead towards the focus. The scattering force pushes the bead in the direction of beam propagation. The trapping position is the equilibrium position where both forces cancel out each other. If the trapping NA is decreased, the forward pushing scattering force is increased relative to the gradient force. This change of the relative radiation pressure shifts the trapping position along the optical axis.



**Fig. 5** Optical forces acting on a trapped bead for high and low trapping lens NAs. The sizes of the yellow and blue arrows indicate the strengths of the gradient and scattering forces respectively.

### 5 Summary and conclusions

We measured the trapping position  $z_t$  of a 1- $\mu\text{m}$  latex bead relative to its interferometric position signal for high (1.20) and low (0.93) trapping lens NAs. We showed that  $z_t$  is shifted along the optical axis and that the unique axial detector range is increased if the NA is decreased. The increased detector range enables 3D tracking in a larger volume. We also measured the trapping position relative to the focus and found that the bead is trapped 140 nm behind the focus for a NA of 1.20. We showed that our observations are in agreement with the two component approach for optical forces.

### References

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