## Supplementary information for 'Sub-diffraction positioning of an optically trapped and two-photon excited quantum dot'

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**Figure S1.** Blue-shift of emission from a two-photon absorption excited QD. **a:** Sketch of the assay, a QD is immobilized on a coverslip and irradiated by the most intense region of the IR laser. **b:** Intensity versus time of a single immobilized QD. The intensity at different emission wavelengths (655 nm, 605 nm, and 565 nm, respectively) are measured with the spectral image splitter. A blueshift of the emitted light starts after  $\sim$ 30-60 seconds of irradiation.

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**Figure S2.** Force measurements of optically trapped QDs. **a:** Corner frequency,  $f_c$ , versus the exit power at the laser for each translational direction. We estimate the on-target power to be 20% of the power. For each data point the corner frequency,  $f_c$ , was found for at least 5 individual QDs. The lines denote best linear fits to data, a linear relation between  $f_c$  and laser power is a hallmark of optical trapping. Also noticeable is the fact that the axial direction, z, is not significantly weaker than the lateral directions, x and y, thus proving a quite well-focused trap in the axial direction. **b:** The spring constant  $\kappa$  of the trap versus the exit power at the laser for each translational direction.  $\kappa = 2\pi\gamma f_c$ , where  $\gamma$  is the friction coefficient and  $f_c$ 's are given in (a).



**Figure S3.** Probability that an optically trapped QD is within a region with at least a certain focal intensity level (black full line), cumulative probability of the QD being two photon excited at a focal certain intensity level (grey full line), and the cumulative probability that the QD is two-photon excited and its emission detected by the EMCCD (dashed black line) at a certain focal intensity level.