Video Article

Optical Trap Loading of Dielectric Microparticles In Air

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Abstract

We demonstrate a method to trap a selected dielectric microparticle in air using radiation pressure from a single-beam gradient optical trap. Randomly scattered dielectric microparticles adhered to a glass substrate are momentarily detached using ultrasonic vibrations generated by a piezoelectric transducer (PZT). Then, the optical beam focused on a selected particle lifts it up to the optical trap while the vibrationally excited microparticles fall back to the substrate. A particle may be trapped at the nominal focus of the trapping beam or at a position above the focus (referred to here as the levitation position) where gravity provides the restoring force. After the measurement, the trapped particle can be placed at a desired position on the substrate in a controlled manner.

In this protocol, an experimental procedure for selective optical trap loading in air is outlined. First, the experimental setup is briefly introduced. Second, the design and fabrication of a PZT holder and a sample enclosure are illustrated in detail. The optical trap loading of a selected microparticle is then demonstrated with step-by-step instructions including sample preparation, launching into the trap, and use of electrostatic force to excite particle motion in the trap and measure charge. Finally, we present recorded particle trajectories of Brownian and ballistic motions of a trapped microparticle in air. These trajectories can be used to measure stiffness or to verify optical alignment through time domain and frequency domain analysis. Selective trap loading enables optical tweezers to track a particle and its changes over repeated trap loadings in a reversible manner, thereby enabling studies of particle-surface interaction.

Video Link

The video component of this article can be found at https://www.jove.com/video/54862/

Introduction

Ashkin reported the acceleration and trapping of microparticles by radiation pressure in 1970.[1] His novel achievement promoted the development of optical trapping techniques as a primary tool for fundamental studies of physics and biophysics.[2,3,4,5] To date, the application of optical trapping has focused mainly on liquid environments, and been used to study a very wide range of systems, from the behavior of colloids to the mechanical properties of single biomolecules.[6,7,8] Application of optical trapping to gaseous media, however, requires resolving several new technical issues.

Recently, optical trapping in air/vacuum has been increasingly applied in fundamental research. Since optical levitation potentially provides nearly-complete isolation of a system from the surrounding environment, the optically levitated particle becomes an ideal laboratory for studying quantum ground states in small objects,[4] measuring high-frequency gravitational waves,[9] and searching for fractional charge.[10] Moreover, the low viscosity of air/vacuum allows one to use inertia to measure the instantaneous velocity of a Brownian particle[11] and to create ballistic motion over a wide range of motion beyond the linear spring-like regime.[12] Therefore, detailed technical information and practices for optical traps in gaseous media have become more valuable to the broader research community.

New experimental techniques are required to load nano/microparticles into optical traps in gaseous media. A piezoelectric transducer (PZT), a device that converts electric energy into mechano-acoustic energy, has been used to deliver small particles into optical traps in air/vacuum[5,12] since the first demonstration of optical levitation.[1] Since then, several loading techniques have been proposed to load smaller particles using volatile aerosols generated by a commercial nebulizer[13] or an acoustic wave generator.[14] The floating aerosols with solid inclusions (particles) randomly pass near the focus and are trapped by chance. Once the aerosol is trapped, the solvent evaporates out and the particle remains in the optical trap. However, these methods are not well suited to identify desired particles from within a sample, load a selected particle and to track its changes if released from the trap. This protocol is intended to provide details to new practitioners on selective optical trap loading in air, including the experimental setup, fabrication of a PZT holder and sample enclosure, trap loading, and data acquisition associated with the analysis of particle motion in both the frequency and time domains. Protocols for trapping in liquid media have also been published.[15,16]

The overall experimental setup is developed on a commercial inverted optical microscope. **Figure 1** shows a schematic diagram of the setup used to demonstrate steps of the selective optical trap loading: freeing the resting microparticles, lifting the chosen particle with the focused beam, measuring its motion, and placing it onto the substrate again. First, translational stages (transverse and vertical) are used to bring a selected microparticle on the substrate to the focus of a trapping laser (wavelength 1064 nm) focused by an objective lens (near-infrared corrected long-working distance objective: NA 0.4, magnification 20X, working distance 20 mm) through the transparent substrate. Then, a piezoelectric launcher (a mechanically pre-loaded ring-type PZT) generates ultrasonic vibrations to break the adhesion between microparticles and a substrate. Thus, any freed particle can be lifted by the single-beam gradient laser trap focused on the selected particle. Once the particle is trapped, it is translated to the center of the sample enclosure containing two parallel conducting plates for electrostatic excitation. Finally, a data acquisition (DAQ) system simultaneously records the particle motion, captured by a quadrant-cell photodetector (QPD), and the applied electric field. After finishing the measurement, the particle is controllably placed onto the substrate so that it can be trapped again in a reversible manner. This overall process can be repeated hundreds of times without particle loss to measure changes such as contact electrification occurring over several trapping cycles. Please refer to our recent article for details.[12]

Protocol

Caution: Please consult all relevant safety programs before the experiment. All the experimental procedures described in this protocol are performed in accordance with the NIST LASER safety program as well as other applicable regulations. Please be sure to select and wear proper personal protective equipment (PPE) such as laser protection glasses designed for the specific wavelength and power. Handling dry nano/microparticles may require additional respiratory protection.

1. Design and Fabrication of a PZT Holder and a Sample Enclosure

1. **Design a PZT holder and a sample enclosure**
   NOTE: Particular design values vary depending on the selection of a PZT.
    1. Open the computer-aided design (CAD) software package. Draw a two-dimensional (2D) sketch of a holder for a given PZT dimension. Develop the 2D sketch to volumetric features using combinations of Extrude/Extrude-cut.
    2. Click Sketch, draw a rectangle and extrude it to make a rectangular cube.
    3. Sketch a disk on the top surface of the cube to define a circularly recessed feature to cover and hold the ring-type PZT.
    4. Define a central hole to have an optical access for both real-time imaging and trapping.
    5. Define a circular guide along the rim of the central hole to insert a flat metallic (copper) ring to concentrate the ultrasonic power toward the center area as shown in **Figure 2a**.
    6. Create two bore holes for M6 screws on the PZT holder to be assembled with a bottom plate (purchased, 4 mm thick bottom aluminum plate with a hole in the center), as shown in **Figure 2c** and **2d**.
    7. In a similar manner, design a rectangular frame of the sample enclosure. Click Sketch, and draw a rectangle, extrude the rectangle to make it a rectangular box.
    8. Draw a smaller rectangle on the top surface of the rectangular box and extrude-cut the rectangle to make it as a rectangular tube.
    9. Draw a smaller rectangle on the side wall of the tube and Extrude-cut to transform it into the frame of sample enclosure box.
    10. Convert these three-dimensional (3D) models into a stereolithography (STL) file format for a 3D printing process (**Figure 2b**).
2. **3D printing of the designed objects**
    1. Open the design file ("-.STL") from the 3D printer operating software. Lay the object flat 0/.and center the object on (0, 0, 0) by clicking the object to select it and using the alignment functions: "Move", "On Platform", and "Center". Orient the PZT holder to face the delicate features upward. The recessed surface will be faced upward.
    2. In the menu go to the "Settings" and the "Quality" tab. Set the printing values as following, Infill: 100%, Number of shells: 2, and Layer height: 0.2 mm.
    3. Preview the objects to check the total print time and make sure the layered objects will be printed as desired. Export the 3D print file in a ".x3g" format and save it to use in the 3D printer.
    4. Turn on the 3D printer and warm it up until the temperature of the extrusion nozzle reaches an operating temperature, 230 °C. Load the design file from a memory card or network drive.
    5. During the warm up, place the Build platform with blue painter's tape to help objects adhere securely. As a thermoplastic material for the printing job, use a polylactic acid (PLA) filament for both objects.
    6. Print the designed objects. Once the printing job is finished, turn off the printer after it has cooled down.
    7. Detach the printed object from the platform using a chisel. Straighten up the printed objects. If the orientation is appropriately chosen, the PZT holder can be directly used without further post-processing.
    8. For the sample enclosure, prepare one pair of indium tin oxide (ITO) coated coverslips and three glass coverslips to cover the frame. Use a diamond cutter to fit the coverslip to the enclosure.
    9. Wire the two parallel conducting plates using a fast drying silver paint to supply voltage across two plates. Glue these five windows onto the sample enclosure using an instant adhesive glue.
       NOTE: The one pair of ITO coated coverslips are installed on the sample enclosure in parallel (facing each other) to provide uniform electric field and to generate ballistic motion of the naturally charged particle along the electric field. The three conventional coverslip cover the rest of sample enclosure surfaces (top and two other sides) to protect the trapped particle from the external flow of air

2. Optical Trap Loading of a Selected Microparticle

1. **Sample preparation**
    1. Store the microparticles in an evacuated desiccator to reduce contact with moisture in the air before the experiment.
    2. Pour out a small portion of microparticles onto a glass slide and immediately put the manufacture's bottle back in the desiccator.
    3. Pick up some of the microparticles with a glass capillary tube. Scatter the particles over the substrate by gently tapping on the capillary while holding the capillary over the coverslip.
    4. Verify the quantity and distribution of deposited particles on the substrate using a dark-field microscope.
       Note: In the sample preparation step, the particle is just scattered on a coverslip and imaged with an optical microscope to verify overall arrangement before inserting them (a coverslip with scattered microparticles) between the PZT and PZT holder. Since the surface

adhesion is strong enough to hold individual microparticles on the substrate, the adhered particles are firmly fixed unless significant external force is applied.

2. **Piezoelectric launcher assembly**
    1. Obtain all the components of the piezoelectric launcher: the flat bottom plate, insulating film, the PZT, the glass coverslip, a copper ring, the PZT holder, two M6 screws, and the sample enclosure.
    2. Apply a thin film (or tape) on the bottom plate to insulate the PZT. The glass coverslip isolates the top of the stack.
    3. Assemble the stack by centering the PZT on top of the flat plate now insulated with tape, followed by the coverslip, the copper ring, and the PZT holder. Screw the stack together maintaining the centering of the PZT to avoid shorting the PZT to the holder if the holder is conducting as shown in **Figure 2c** and **2d**. The copper ring provides an evenly distributed mechanical preload on the stack for plastic PZT holders.
    4. Finally, glue the sample enclosure onto the stack and mount the assembly on an XYZ translational stage in the microscope.

3. **Configuration of the PZT launcher**
NOTE: Driving the PZT with a high voltage signal has potential electrical hazards. Please consult with safety personnel before the experiment. All the electrical connections should be secured before the experiment. Turn off the amplifier and disconnect PZT leads whenever possible.
    1. Connect the PZT leads to the voltage amplifier and connect the function generator to an input port of the voltage amplifier.
    2. Turn on the function generator and configure it to generate continuous square waves with an output voltage of 1 V. Do not generate the voltage signal until all the connections are verified and secured.
    3. Turn on the voltage amplifier and generate the square wave of output voltage 1 V by enabling the output.
    4. Connect the monitoring output port (output voltage 200 V) of the amplifier to an oscilloscope. Configure the amplifier to have gain of 200 V/V by turning the gain knob on the front panel. Verify that the monitoring output voltage has an amplitude of 1 V as measured by the oscilloscope.
    5. Once the function generator and the amplifier are configured, find the resonant frequency of the PZT launcher by scanning the modulation frequency of the driving signal while the real-time video microscope images adhered particles. Repeat the scanning until the microparticle motion is a maximum. Use this frequency (64 kHz here) to release particles.
    NOTE: The modulation frequency is manually changed (scanned) from zero to 150 kHz to find the resonant frequency.
    6. Configure the function generator to generate a square wave with a specified number of cycles in burst mode. Press the "Burst" button on the front panel and select "N Cycle Burst".
    7. Choose the burst count by pressing "# Cycles" soft key and set the count to 10 or 20.
    8. Configure the square waveform to generate voltage signals with an amplitude of 600 V (three times the voltage used for continuous excitation) at the resonant frequency of 64 kHz which has found from the previous step. Verify that the pulsing signal releases the target particle in a repeatable manner by ensuring particles move after each pulse.

4. **Selective optical trap loading**
NOTE: The PZT launcher assembly is installed on a manual linear translation xy stage. The particles can be translated relative to the fixed beam focus by moving the translational stage.
    1. Remove the laser line filter to identify the focus of the trapping beam by rotating the microscope turret (**Figure 3a**). Move the motorized focusing block back and forth vertically around the best focus of the visible image to optimize focus.
    2. Once the focus position is verified, put the filter back to give a clear real-time video without interference from the trapping beam.
    3. Translate the sample to place a selected particle at the focus position of the trapping laser. Focus on the particle to image the center of a selected particle, which places the nominal trapping position below the particle center by about one half radius while leaving the levitation position above the particle.
    4. Adjust the power supply connected to the electro-optic modulator (EOM) driver to set the optical trapping power. The optimal power depends on particle size and material. The optical power was found through repeated trials to determine the power sufficient to levitate the particle without ejecting it from the beam. Here, use an optical power of 140 mW at the back focal plane of the objective to trap the 20 µm diameter polystyrene (PS) particles.
    5. After the center of the selected particle is aligned, actuate the piezoelectric launcher with several pulses. The change of the particle image from a static focused image to a moving blurred image indicates successful loading to the levitation position.
    6. Translate the levitated particle vertically about a millimeter above the substrate by moving the objective lens up to prevent possible surface interactions. Then reduce the optical power to transition the levitated particle (**Figure 3b**) into the nominal trapping position (**Figure 3c**) which is more stable.
    NOTE: The optical power of trapping laser can be modulated by an electro-optic modulator (EOM). The EOM regulates the output power with a bias voltage supplied through a digital power supply. One can observe the transition from the levitation to trapping position through the CCD while slowly reduces the optical power.
    7. For the position measurement, as depicted in **Figure 3c** to **3d**, carefully move the center of the PZT holder to the optical axis and then move the objective lens up (vertically) to translate the particle into the middle of sample enclosure (9 mm above the substrate) where the fringe electric field is minimized.
    8. After performing the measurement as described below, place the particle on the substrate by moving the objective down until the particle touches the substrate. Since most of the particles are applied near the corners, the trapped particle can be easily recognized and re-trapped when it is placed in the central area. This enables reversible trap loading to measure changes occurring beyond a single trapping event such as contact interactions of the particle and substrate.

3. Data Acquisition

1. Align the condenser and the focusing lens to maximize the QPD "SUM" signal with a particle in the trap.
2. Align the focusing lens to nominally zero the X and Y channels of the QPD, as shown in **Figure 4c**.

3. Repeat the adjustment of the condenser and the focusing lens until the Fourier transformed position signals (or power spectrum density (PSD) plots) of the X and Y channels superimpose to show balanced sensitivity. Properly aligned QPD signals (X and Y) show almost identical behavior, as shown in **Figure 4b**.
4. Once the QPD alignment is verified, connect the voltage amplifier to the two ITO plates. Connect the voltage monitoring output signal of the amplifier to the DAQ system to record the step excitation signal and the induced particle trajectory synchronously.
5. Supply a continuous square wave of 400 V to generate an electric field (**Figure 4d**) that moves the particle transversely to the optical axis by about 500 nm (**Figure 4e**). Measure the step response of the trapped particle using the QPD.
6. Average multiple periods as necessary to reduce the effects of Brownian motion. The induced motion can be used to measure the optical force over a wider range of motion than that of thermal fluctuations.[12,17] **Figure 4d** and **4e** shows averaged signals of applied voltage and the induced particle trajectory over 50 iterations of step excitation.

Representative Results

The PZT launcher is designed using a CAD software package. Here, we use a simple sandwich structure for the preloading (a PZT clamped with two plates), as shown in **Figure 2.** The PZT holder and the sample enclosure can be fabricated from a variety of materials and methods. For a quick demonstration, we choose 3D printing with thermoplastic as illustrated in **Figure 2d**. Based on the fabricated components, optical trap loading is shown in **Figure 3**. For selective loading, the reflected trapping laser is blocked during the experiment by a filter installed on a microscope turret to protect the CCD camera while the visible light passes the filter for imaging in reflection as illustrated in **Figure 1.** A calibrated CCD camera also facilitates quantitative measurement by allowing measurement of the particle diameter and additional position detection. The diameter of a target particle can be used to calculate the mass which yields trap stiffness from the natural frequency, as discussed below. The trajectories measured using the CCD camera are also used to calibrate the QPD voltage signal for measuring the displacement.[12]

Once the particle is trapped, bright scattering from a red laser allows the trapped particle to be recognized with the naked eye, as shown in **Figure 1** (inset photograph). Also, real-time images of the substrate can determine if the particle has been trapped since it is at a different height (focus) from the other microparticles adhered to the substrate (**Figure 3**). The microparticles can be trapped in two positions: a trapping position and a levitation position. In the trapping position, optical forces stabilize the particle in all directions. In contrast, in the levitation position the particle is only stabilized transversely by optical forces. In the vertical the upward force from radiation pressure is balanced by gravity. With our loading method, the selected particle is generally delivered to a levitation position. At the levitation position, the vertical location of the suspended particle is much more sensitive to variations in the optical power than at the trapping position near the focus.[18] One can vertically move the particle repeatably between these two stable positions by varying the optical power. The levitation position also has higher sensitivity to external forces than the nominal trapping position because the trap stiffness becomes softer as the light propagates away from the focus. Therefore, the levitation position can also be used for more sensitive measurements when displacement noise is not dominated by brownian motion. When the position noise is thermally limited as it is here, decreasing the stiffness increases both sensitivity and noise so there is no gain for precision measurement.

The motion of the trapped particle is monitored by a QPD and recorded by a DAQ board. The QPD signal is recorded in the time domain (**Figure 4c**) and Fourier transformed (**Figures 4a** and **4b**). The overall alignment can be conveniently checked by comparing the power spectra of two radial channels (X and Y). If they are not superimposed (**Figure 4a**), the optical alignment has to be corrected until superposition occurs (as shown in **Figure 4b**).

The particle trajectory shows both Brownian and ballistic motion as shown in **Figure 4**. Time and frequency domain analyses can be used to interpret these measurements. We have introduced two approaches to force measurement which allow more complete understanding of the optical trap by comparing Brownian motion to the Ballistic motion induced by an electrostatic force. The particle trajectory for Brownian motion under no electrostatic field is converted to the power spectral density which can then be analyzed by a nonlinear least square fit the solution of the full Langevin equation.[19] This analysis of the PSD yields the resonant frequency and damping near the trap center. The resonant frequency is converted to the trap stiffness using the known mass in the formula $\omega_o = \sqrt{k/m}$. The measured displacement then gives the optical force using the formula for a spring $F = -kx$.

The ballistic motion induced by a step change in the electrostatic field can also yield the resonant frequency of the trap and damping of the medium.[12] As we remove the electrostatic field from the trapped particle, the particle will be released to return to the field-free tapping position.as shown in **Figure 4d** and **4e**. The displacement as function of time can be fit to the general solution of a damped harmonic oscillator to give the resonant frequency, damping, and steady-state displacement. Both of these approaches assume that the particle in the trap acts as a linear spring. These measurements can be extended to general (non-linear) forces using the parametric force method.[12] The details of the PSD analysis and parametric force analysis are not the focus in this protocol but they can be found from the literature.[12,19]

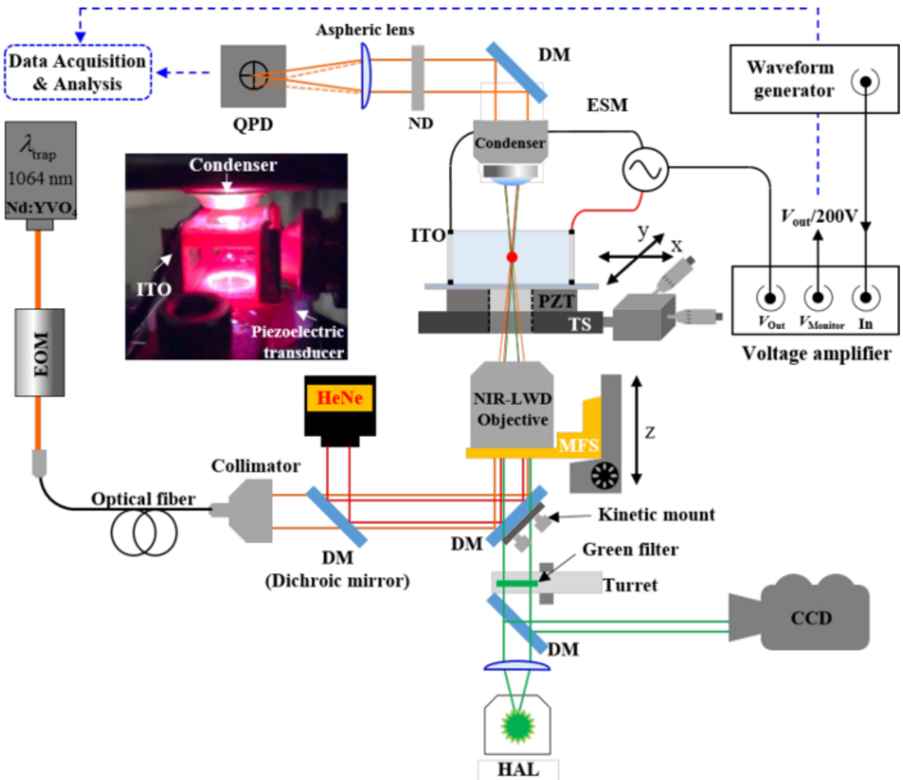

Figure 1: Schematics of the Experimental Setup used for Selective Optical Trap Loading in Air. A single-beam gradient force optical trap is developed on an inverted optical microscope. Abbreviations used in the schematic are listed below: EOM, electro-optic modulator; HAL, halogen illuminator; MFS, motorized focusing stage; NIR-LWD objective, infrared corrected long working distance objective lens; TS, translation stage (x −y); PZT, piezoelectric transducer; ESM, electrostatic field modulator; ND, neutral density filter; QPD, quadrant-cell photodetector; DM, dielectric mirror; ITO, indium tin oxide coated coverslips; CCD, charge coupled device camera; HeNe, helium neon laser (633 nm); Nd:YVO_4, 1,064 nm laser for trapping.[12] Please click here to view a larger version of this figure.

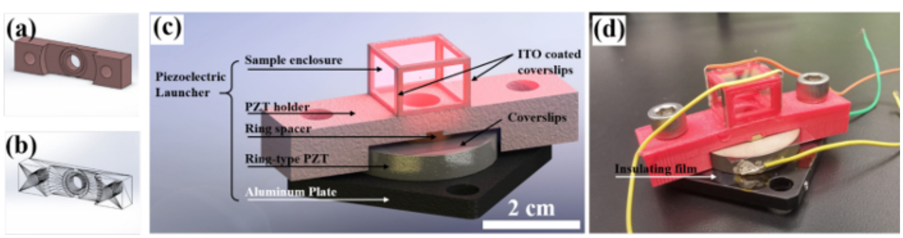

Figure 2: Fabrication of the Piezoelectric Launcher Assembly. (**a**) Rendered images of a PZT holder using CAD software package in a "-.SLDPRT" format and (**b**) "-.STL" format for 3D printing. (**c**) A rendered image of the final assembly of the piezoelectric launcher: sample enclosure (with ITO coated coverslips), PZT holder, ring spacer, ring-type PZT, aluminum plate, coverslips. (**d**) Picture of the final assembly. Please click here to view a larger version of this figure.

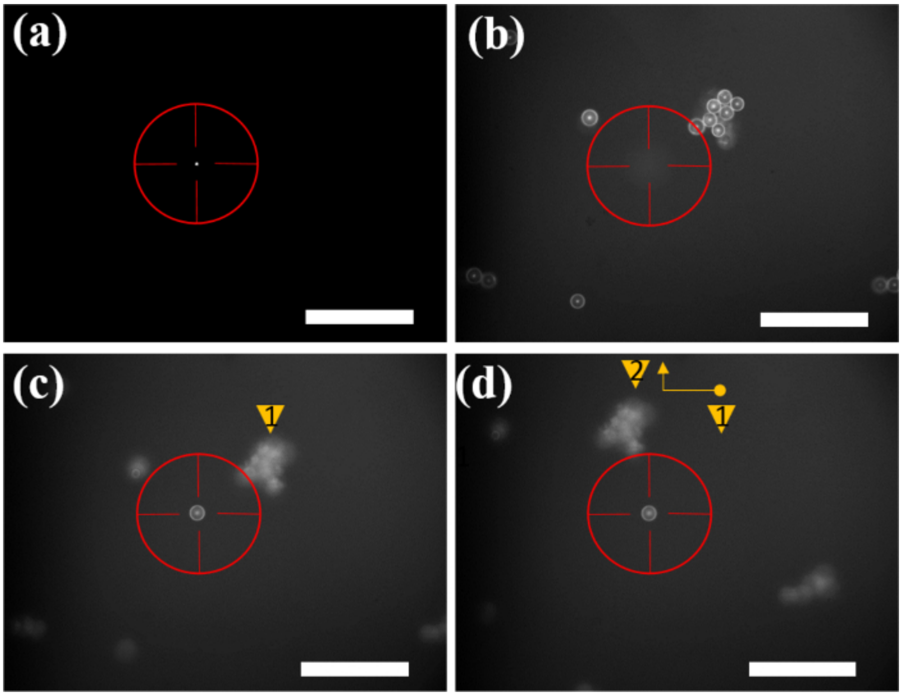

Figure 3: Step by Step Demonstration of Selective Optical Trap Loading of a 20 µm PS Particle. (**a**) locating the focus of the trapping beam, (**b**) levitating the particle above focus (The particle image is a dim blur because the levitation position is well above the nominal microscope focus), (**c**) transitioning into the trapping position (nominally in focus), and then (**d**) moving the trapped particle to the central area for data acquisition. The particle is trapped at a fixed location of the beam focus whereas the sample stage is moved as indicated with a yellow arrow in Figure 3d (Scale bar = 100 µm). Please click here to view a larger version of this figure.

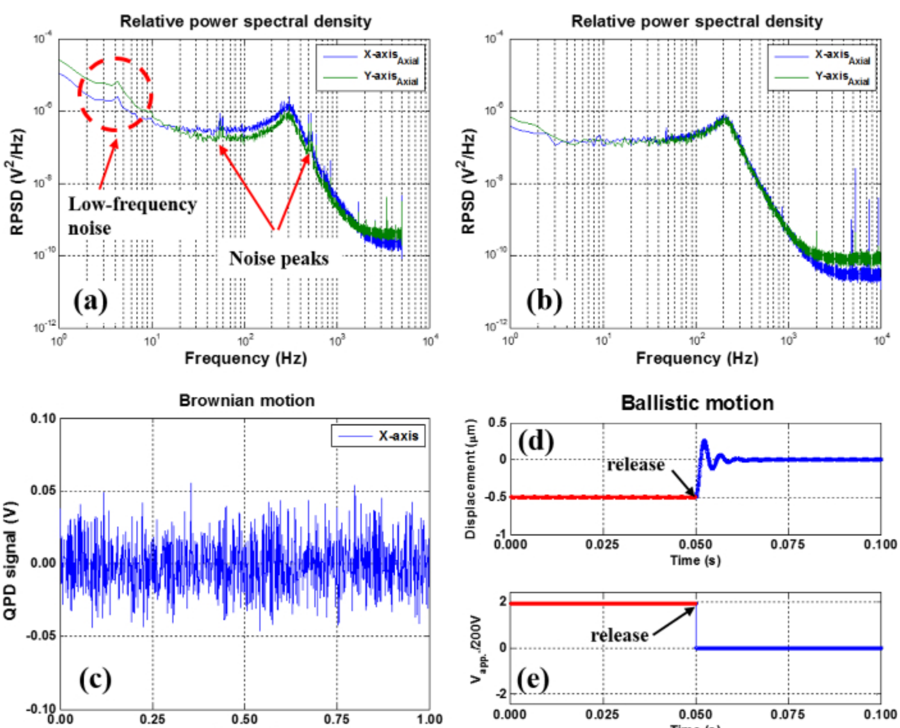

Figure 4: QPD Captured Particle Trajectories Both in Frequency and Time Domain. (**a**) A poorly aligned experimental setup shows low-frequency noise and noise peaks at specific frequencies whereas (**b**) well-matched PSDs of the x and y-axis indicate correct optical alignment. (**c**) A QPD records the Brownian motion of the trapped particle in the time domain. (**e**) A step change in applied electric field across the trapped particle is synchronously recorded with the induced (**d**) ballistic motion through the data acquisition (DAQ) system. Please click here to view a larger version of this figure.

Discussion

The piezoelectric launcher is designed to optimize the dynamic performance of a selected PZT. Proper selection of PZT materials and management of ultrasonic vibrations are the key steps to yield a successful experiment. PZTs have different characteristics depending on the type of transducer (bulk or stacked) and component materials (hard or soft). A bulk type PZT made of a hard piezoelectric material is chosen for the following reasons. First, hard piezoelectric materials have lower dielectric losses and higher mechanical quality factor than soft materials. Second, the bulk type PZT represents a lower electrical load and is easier to drive at high frequencies than a stacked type transducer. Under dynamic operation, high amplitude oscillation can cause tensile forces on an unloaded PZT ceramic that result in mechanical failure. A mechanical preloading structure is used to provide a constant load to reduce backlash and enhance dynamic performance of the PZT. A metallic ring spacer is inserted between the PZT holder and the ring-type PZT. This metallic ring spacer concentrates the ultrasonic power and distributes it evenly around the ring (Any local (uneven) stress can easily break the coverslip.). With a well-designed PZT launcher, proper alignment of the particle to the trapping beam in both axial and radial directions determines the efficiency of trap loading. If the particle is not successfully levitated after pulsing, repeat the substrate alignment and move the focus a little below the particle to find the optical loading position. For the near-infrared corrected objective lens, the focus of the trapping beam is set to be a few micrometers below the sample plane that is focused onto the CCD. The optimal trapping power required to trap microparticles varies as the size of the target microparticle changes.[13] The optimal trapping power can be found empirically through trial and error. The power required here (140 mW) is relatively high due to the low NA and long working distance used.

Here we demonstrated reversible trap loading of a 20 µm PS particle. However, our approach can be extended to smaller particles. For smaller microparticles, our current PZT launcher may not able to provide enough ultrasonic power to detach the particles. Use of a faster PZT driving circuit has been shown to release smaller particles.[20] In addition, a low-adhesion surface can be an alternative approach.[21] Reduction of the adhesion between microparticles and the substrate will mitigate the minimum ultrasonic power required to detach the particle thus our current PZT launcher can also be used to detach smaller particles.

Most conventional loading techniques are random processes in which numerous aerosol droplets with solid inclusions are continuously generated until one of them is trapped by chance near the trap center. Thus this conventional technique may not be appropriate for trapping samples with a limited quantity or maintaining uniform sampling. In the protocol, we demonstrate reversible optical trap loading which includes repeated cycles of trap loading and landing. This enables unique experiments, for example the study of charge accumulation on the particle.[22] The charge on the trapped particle can be measured by fitting the transient response (**Figure 4d**) to the ideal solution of harmonic oscillator in a nonlinear least square manner. The induced displacement multiplied by trap stiffness gives the electrostatic force which allows calculation of charge from the known electric field strength (given by the applied voltage divided by the distance between the two parallel ITO coated plates).[12] This simple charge measurement can be extended to study particle-surface interaction when combined with the reversible trap loading technique demonstrated here.[22]

Disclosures

The authors declare no competing financial interest.

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