Determining the Stiffness of an Optical Trap

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In this report we will examine the stiffness of the laser, κ, used in an optical trap. The results are obtained through fitting a Gaussian equation,

$$\rho(x) = \frac{1}{\sqrt{2\pi}\sigma} \exp\Big[-\frac{(x-x_0)^2}{2\sigma^2}\Big] \tag{1}$$

to the probability density histogram of our data, where $\sigma^2 = \frac{\kappa}{k_B T}$ and k_B is the Boltzmann constand and T is the temperature assumed to be room temperature. Then the value of κ is found from the σ from the fit.

The experimental setup consists of a combination of two other setups: a microscope setup and a laser setup. The microscope setups consists of an XYZ translator, a 25mm lens, a 50mm lens, an objective lens, a CCD camera[1] and a sample containing the beads that we will analyze the motion of. The laser setup is made of a laser, a lens and two mirrors to control the degrees of freedom and a dichroic mirror. The microscope portion works by focusing a beam of light on the sample with the two lenses and then projecting that image with objective lens onto the CCD camera. The laser trap is added by using the mirrors to redirect the laser into the rear of the objective lens and focusing on the sample, thus creating a trap. Then a freely moving bead is trapped and the data of its motion is collected for a few minutes at around 66 data points per second.

Fig 1 shows the raw data of the position of the bead in the x axis while being trapped by the laser. As the actual data is not centred around zero, the mean of the data was calculated and found to be 441px, and each data point has the mean subtracted from it to centre is at zero. The conversion factor from pixel to μm for this particular camera is 3.45μm per pixel. For this set of data, the lens used to focus the laser was a 100mm lens and it was placed 26cm behind the rear of the objective lens to focus it. The distance was measured by measuring from the lens of the objective to the middle of the 100mm lens. It can be seen in our data that the larger shifts in position are consistently in the negative x direction which indicates that there was some factor causing a slight bias towards one side. As the number of shifts is small, we took close to 10^4 to make sure that the shift is negligible. Overall, the data stays within 1 or 2 pixels of the mean which indicates the effectiveness of our trap. Fig 2 shows the raw data of the position of the bead in the y axis which once again shows a bias towards the positive y axis in its extremities. Fortunately taking a large amount of points also made this negligible as it will be seen when creating a histogram of the data.

Fig 3 shows the histogram of the x position data normalized to a probability density function. It can be seen here that the large fluctuations are now near zero and insignificant compared to the data centred around the mean. The error for this histogram is $\sqrt{N}$ of each bin divided by $N\Delta x$. The position of the bead in the x axis seems to largely be within the -1 to 1 pixel range.

Fig 4 shows the fit of the data from Fig 3 using Eq. 2 weighted to the the error along with its residuals. . The fit has a χ^2 value of value of 67.5 which is reasonable due to several points having a weight of zero. The σ received from the fit was 0.773±0.013 $\frac{}{\text{px}}$ which leads to a κ value of $(1.7 \pm 0.2) \times 10^{-6}$ N/m when converted to N/m units. The fit was also done with the y data and it was found that σ was 0.74±0.09 $\frac{}{\text{px}}$ and has a κ value of $(1.8 \pm 0.2) \times 10^{-6}$ N/m when converted to N/m units and taking into account that the image is magnified by 100.

For comparison, the Monte Carlo method was used to simulate a data set of 10^5 points and it was fit the same way the original data was fit. This simulated data gave a κ of $(3.5 \pm 0.4) \times 10^{-6}$ N/m which somewhat agrees with our data. Another way to calculate a $1/\sigma$ value was from the variance of the data using the equipartition theorem

$$\frac{1}{2}\kappa < x^2 >= \frac{1}{2}k_B T \tag{2}$$

which resulted in a κ of $(6.1 \pm 0.5) \times 10^{-5}$ N/m. The value from the equipartition theorem is off by a factor of 10 compared to the previous results while the simulated data and the actual data somewhat agree with each other.

The results received from the experiment are not quite as expected as in the background file for this experiment it was mentioned that κ should be around 10^{-5} N/m and the value calculated was one order of 10 from what was expected. This error could be the result of the size of the laser when it focuses on the sample and it was quite larger than the bead which could have affected the strength of the trap. Some other sources of error in the experiment could be that the drift in the x and y axes was more significant to the plot that expected, and shifted affected the fit for the data. The drift could have resulted from a sample that was leaking or from a fault in the setup.

[1] FLIR Blackfly UFS-U3-16S2M camera. See https://www.ptgrey.com/blackfly-s-mono-16-mp-usb3-vision-sony-imx273.

FIGURES

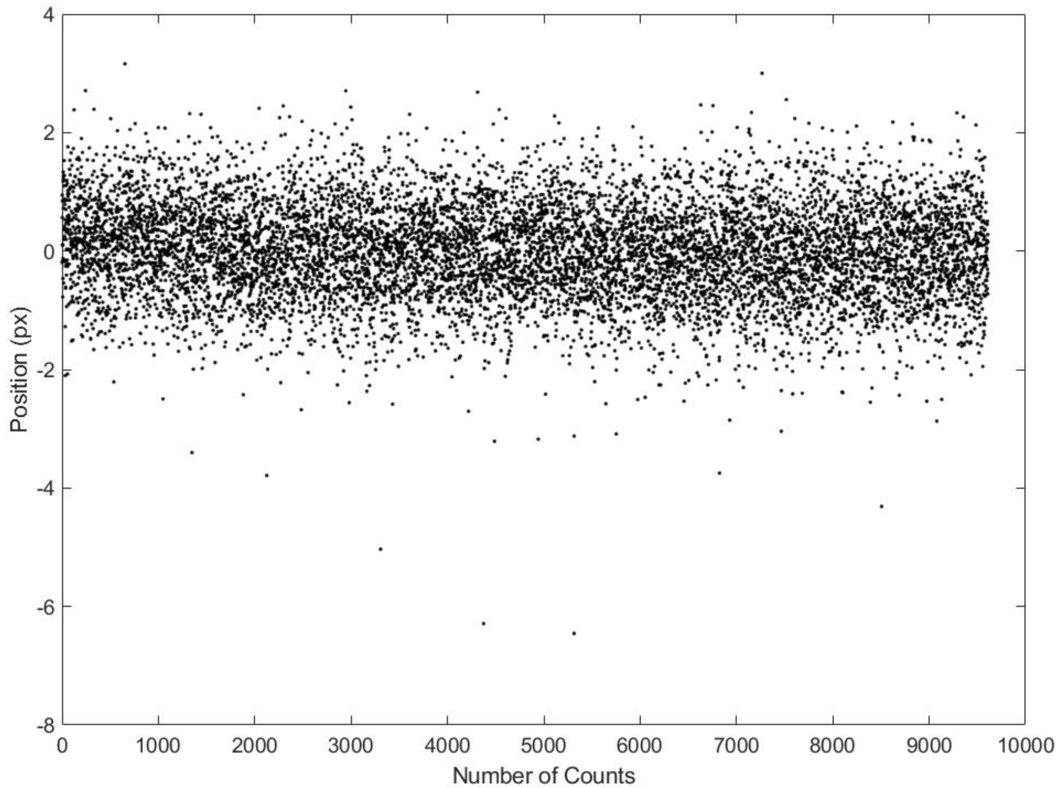

FIG. 1. A plot of the raw data of the displacement of the bead in the x axis. The data is subtracted by the mean of all data points to centre it around 0.

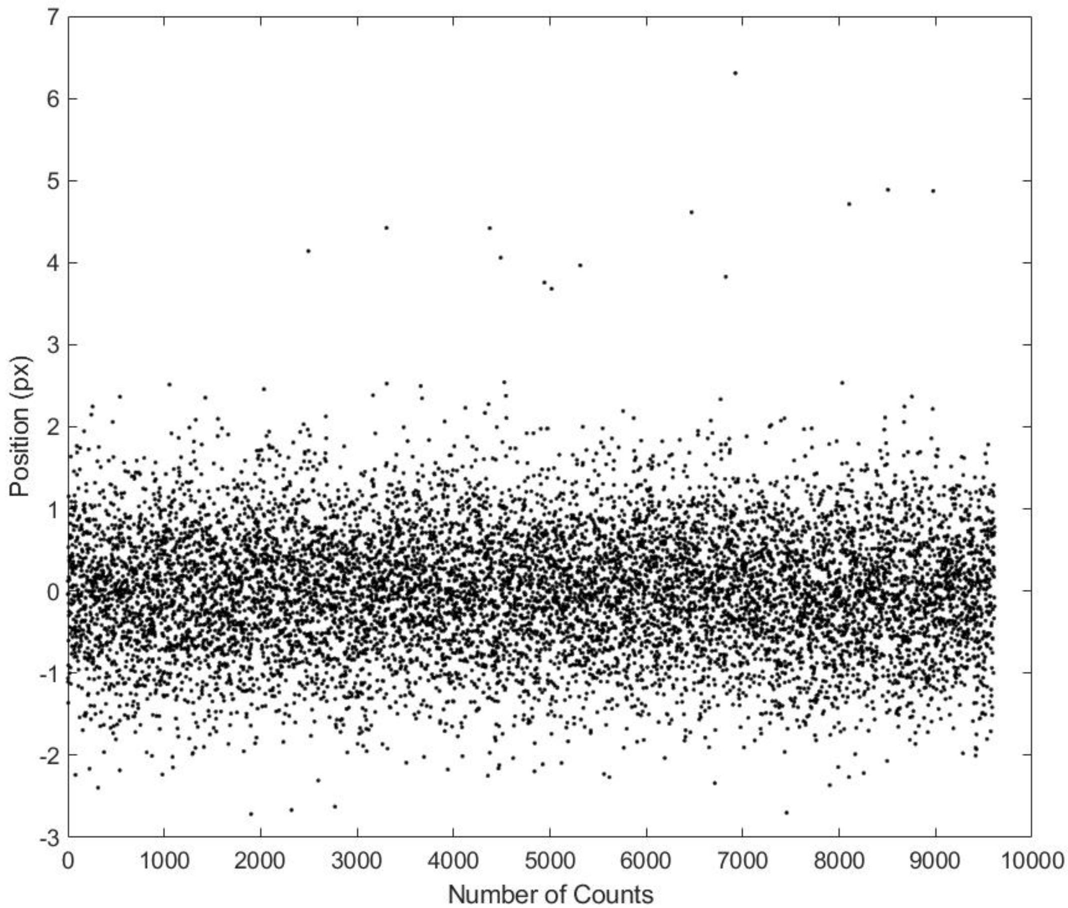

FIG. 2. A plot of the displacement of the bead in the y axis. The data is subtracted by the mean of all data points to centre it around 0.

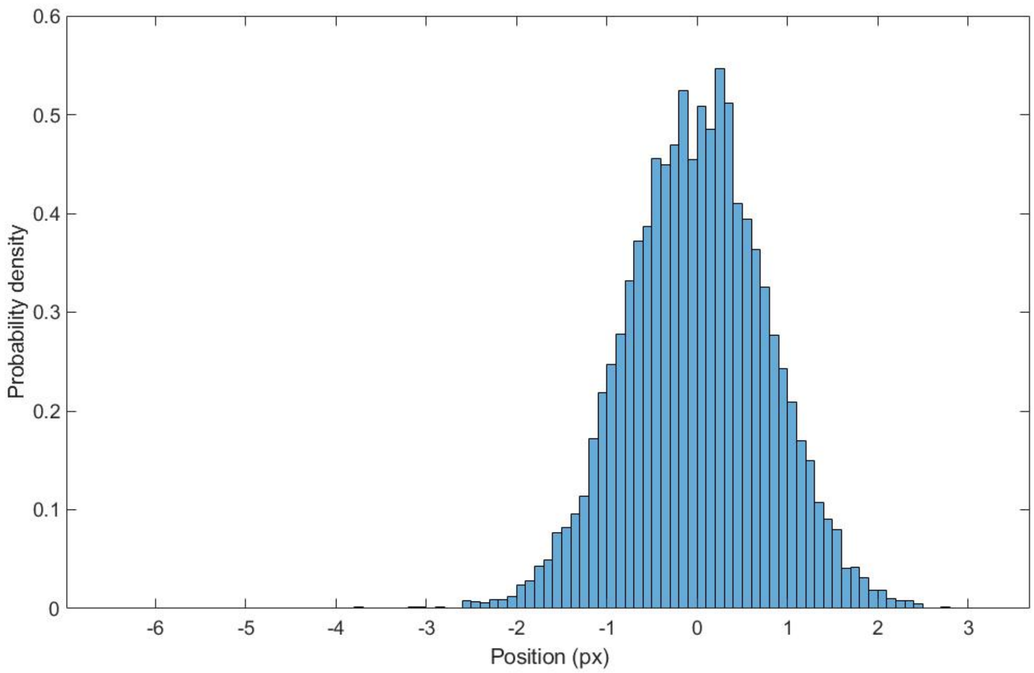

FIG. 3. A histogram of the data of the displacement of the bead in the x axis normalized to a probability distribution function.

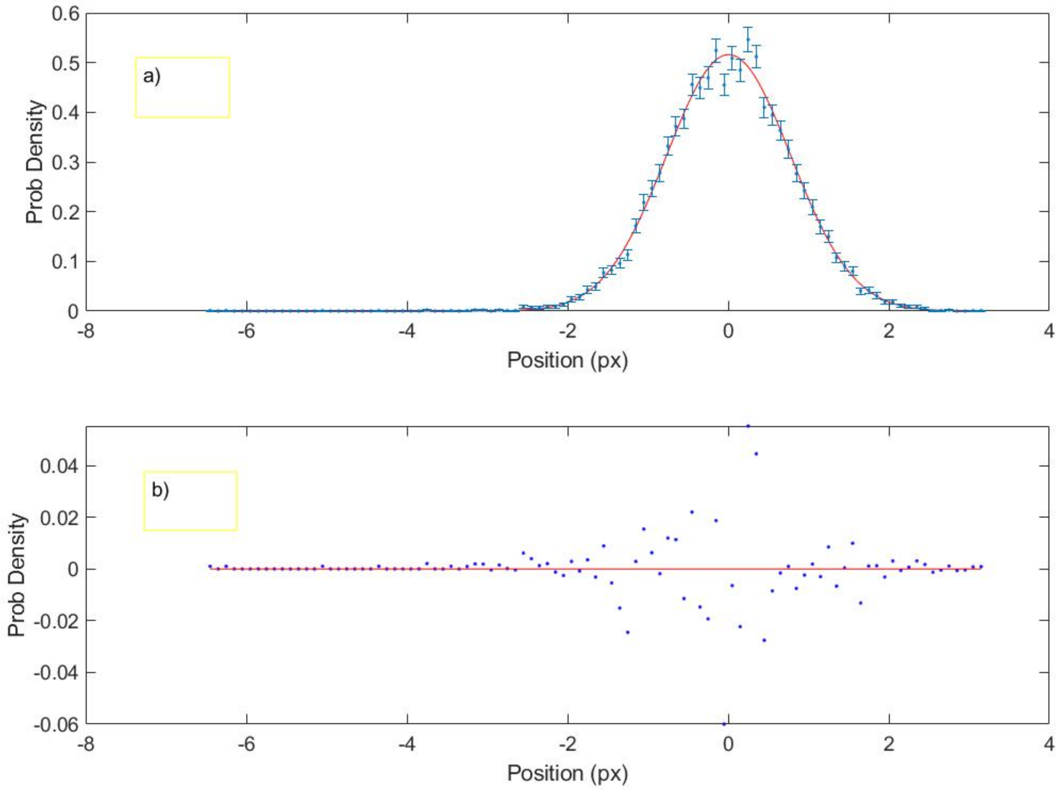

FIG. 4. Two plots containing the fit of the probability histogram data to Eq 2 and its residual data. a) is the plot of the data obtained from Fig 3 fitted to Eq 2. The weights of the fit are from the errors of the histogram normalized to a probability density function. b) is the plot of the residuals of the fit on a).