Evanescent light scattering Optical Tweezers Random Walk

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Abstract auf Englisch (10-15 Zeilen) Hello, here is some text without a meaning. This text should show what a printed text will look like at this place. If you read this text, you will get no information. Really? Is there no information? Is there a difference between this text and some nonsense like "Huardest gefburn"? Kjift – not at all! A blind text like this gives you information about the selected font, how the letters are written and an impression of the look. This text should contain all letters of the alphabet and it should be written in of the original language. There is no need for special content, but the length of words should match the language. Hello, here is some text without a meaning. This text should show what a printed text will look like at this place. If you read this text, you will get no information. Really? Is there no information? Is there a difference between this text and some nonsense like "Huardest gefburn"? Kjift – not at all! A blind text like this gives you information about the selected font, how the letters are written and an impression of the look. This text should contain all letters of the alphabet and it should be written in of the original language. There is no need for special content, but the length of words should match the language.

Available at www.github.com/leoole100/fp2.

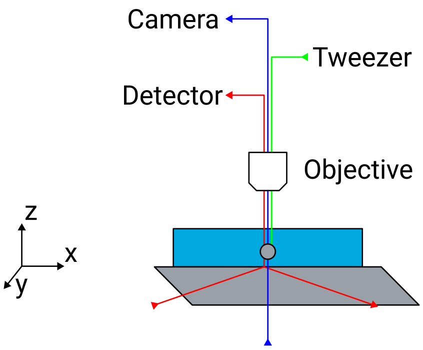

Figure 1 Schematic of the experimental setup. Optical tweezers in green, transmission light microscopy in blue and total internal reflection microscopy in red.

1 Introduction

Using the statistics of the random walk small forces can be measured. This is demonstrated on colloidal particles in a aqueous solution. Forces are applied using optical tweezers and the position is measured with different microscopy techniques.

2 Methods

This experiment operates with light on small particles (in the range of 1 µm [1]) in a aqueous solution. This is shown in Figure 1 as the gray circle in the blue box.

Using a optical tweezers setup the particle can be trapped in the center of the experiment. This results on a 3 dimensional harmonic potential, with a spring constant that is roughly proportional to the trap strength.

To directly observe the particle transmission light microscopy is used. For this a blue led is used to illuminate the particle from below. This allows the particle position to measured in x and y direction.

To measure the position in z direction, total internal reflection microscopy is used. For this a red laser is bounced off a prism to create a evanescent wave in the aqueous solution. This wave scatters off the particle into a sensitive photo detector.

3 Results

All recorded data and the analysis is available at www.github.com/leoole100/fp2.

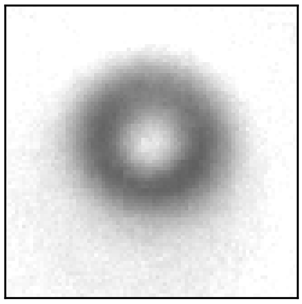

Figure 2 Transmission light microscopy of a observed particle. The radius of the particle is 14(2) px, equivalent to 1.86(27) µm.

3.1 Transmission Light Microscopy

In the first section of the experiment, the particles were observed using a transmission light microscope setup. For this images with a resolution of 600 × 800px were recorded with a frequency of 10 Hz for 10 min. The magnification of the microscope was assumed to be 0.133 196 72 µm/px [1], this is the main systematic error of this measurement procedure.

The images were normalized with a black (illumination off) and white (illumination on, particle not in frame) reference image, to remove the influence of dust in the imaging elements. The particle that was used for this experiment is shown in Figure 2, after the normalization.

To determine the trajectory of the particle, a effective center of mass was calculated for each frame:

$$\vec{r}(t) = \iint \vec{r} \cdot \left(1 - I(\vec{r}, t)\right)^2 d\vec{r} \qquad (1)$$

The density of the resulting trajectory is shown in Figure 3 for different optical trap stiffnesses. The approximate radius of the optical trap of 2.5 µm is drawn as a red circle. For the trap stiffness of 0, the particle is free to wander around, for the higher stiffnesses like 1.01 the particle is mostly confined to the trap.
For a weak trap like 0.75 the particle is still mostly confined to the trap, but can escape the linear trap region and randomly wander around. This happened multiple times during the shown measurement.

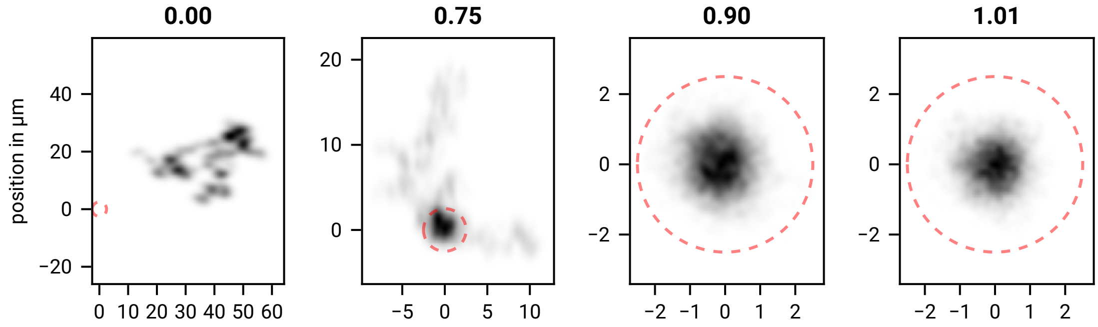

Figure 3 Density of recorded particle positions, grouped by optical trap stiffness. The red circle indicates the approximate radius of the optical trap 2.5 µm.

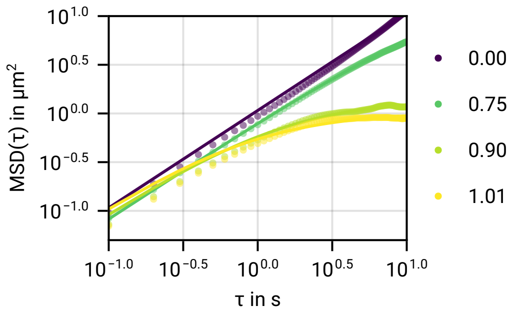

Figure 4 Mean Square Displacement for different optical trap stiffnesses, fit: Equation 3

Mean Square Displacement

A method to describe the trajectory of a random walk is the mean square displacement (MSD) [2]. This is defined as the average of the squared distance of the particle from the starting point [2]:

$$\mathrm{MSD}(t) = \frac{1}{N}\sum_{i}^{N}\left(x_i(t) - x_i(0)\right)^2 \tag{2}$$

Here a different implementation using the autocorrelation of the velocity was used, to achieve a more stable result. This was implemented by [3]. The resulting MSD for different optical trap stiffnesses is shown in Figure 4.

The MSD can be described by the following model:

$$\mathrm{MSD}(\tau) = \frac{1}{\frac{1}{D_0\tau} + \frac{1}{\mathrm{MSD}(\infty)}} \tag{3}$$

For a free particle the $\mathrm{MSD}(\infty) = \infty$ and the MSD grows linearly with D_0 over time [1, 2].
The through the fit estimated diffusion Coefficient D_0 in Table 1 is equivalent with the expected value

Stiffness	D_0 in µm²/s	$k_{\mathrm{MSD}(\infty)}$	k_V
0	0.107 28(93)	NA	0
0.75	0.077 93(3)	NA	0.9011(45)
0.90	0.0678(24)	6.471(62)	4.7177(62)
1.01	0.053 84(8)	8.968(23)	6.021(12)

Table 1 Estimated diffusion constant and spring constant in nN/m.

of $D_0 = \frac{k_B T}{6\pi\eta r} = 0.122(18)\,\mu\mathrm{m}^2/\mathrm{s}$ [1], with $r = 1.86(27)\,\mu\mathrm{m}$ and $\eta = 0.955\,\mathrm{mPa\cdot s}$[4]. The error in the magnification of the microscope does increase the uncertainty.

For a confined particle the MSD reaches a plateau at $\mathrm{MSD}(\infty)$ [1]. This happens for the higher trap stiffness (0.90, 1.01) and the estimated values $k_{\mathrm{MSD}(\infty)} = \frac{2k_B T}{\mathrm{MSD}(\infty)}$ are shown in Table 1 and Figure 6.
For the lower measured spring stiffness (0.75), the MSD does not reach a plateau, as it partially escapes the trap and wanders around (see Figure 3). Therefore this procedure is not adequate for such low spring stiffnesses.

Potential

A more detailed analysis can be done by looking at the distribution of the particle positions. For this the probability density function (PDF) of the particle positions has to be estimated. This is done using the kernel density estimation (KDE) with a Gaussian kernel [5]. The resulting 2 dimensional PDF is shown in Figure 3.
As the density is small for large parts of the explored 2d space, the data is reduced by aggregating along the image coordinates x and y. As the Potential should by rotationally symmetric, the

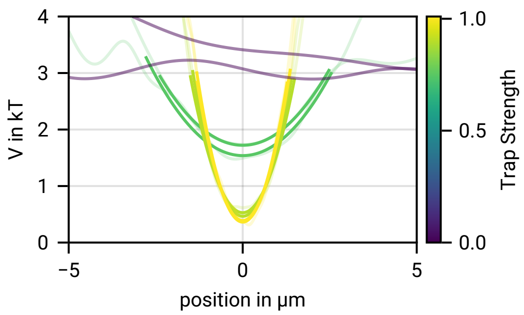

Figure 5 Measured Potential, with quadratic fit.

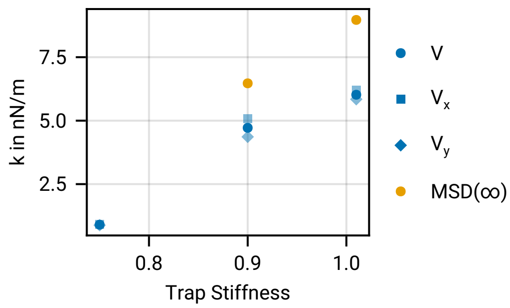

Figure 6 Differently measured spring constants.

data from the x and y axis should not significantly deviate.

Using the Maxwell Boltzmann relations [1], the potential at a position p can be calculated from the PDF:

$$V(p) = -\frac{\log \mathrm{PDF}(p)}{k_B T} \tag{4}$$

The resulting potential is shown in Figure 5. The parts of the measurements with a $\mathrm{PDF}(x) > 0.05$ are used for a quadratic fit, to estimate the spring constant. The resulting spring constants are shown in Figure 6 and Table 1 and are in the same order of magnitude.

For the lower spring constant of 0.75, the potential begins quadratic, but flattens after approximately 2.5 µm. This is due to the limited size of the trap, but the spring constant can still be estimated.

This method allows for the measurement of a spring constant as low as 0.9011(45) nN/m, which means over the used length scales that forces in the order of 1 fN can be measured.

3.2 Total Internal Reflection Microscopy

As references in [1] led to nowhere, we implemented our own methods.

4 Discussion

References

[1] *Evanescent light scattering experiment — Instructions*. Forgeschrittenen Praktikum — University Konstanz. 2023.

[2] Wikipedia. *Mittlere quadratische Verschiebung — Wikipedia*. 2023. URL: https://de.wikipedia.org/w/index.php?title=Mittlere_quadratische_Verschiebung&oldid=234078066.

[3] Riccardo Foffi. *MeanSquaredDisplacement.jl*. 2023. URL: https://github.com/mastrof/MeanSquaredDisplacement.jl.

[4] Anton Paar. IAPWS. 2008. URL: https://wiki.anton-paar.com/en/water/.

[5] JuliaStats. *KernelDensity.jl*. 2024. URL: https://github.com/JuliaStats/KernelDensity.jl.