

# Introduction

The purpose of this website is to explain how to extract information on the heterogeneity of a system from multiple particle tracking data. We begin with a brief overview of multiple particle tracking (MPT) and microrheology before moving on to the topic of heterogeneity. More extensive introductions to MPT can be found in the paper by Crocker & Grier (1996) and the website "Particle tracking using IDL", both referenced [here](#).

What is multiple particle tracking?

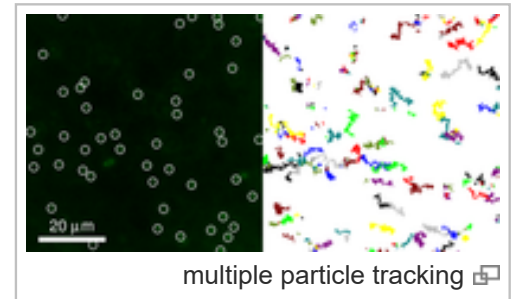
How do the data look like?

An important quantity: the mean-squared displacement (msd)

Microrheology

## What is multiple particle tracking?

Multiple Particle Tracking is a technique that allows you to simultaneously track several micron-sized particles using video microscopy. You record video microscopy movies of microspheres undergoing Brownian motion, and using the image processing software given on the website "Particle tracking using IDL" [linked here](#), you can extract from the movies the individual trajectories of the particles. This is illustrated in the movie on the right (click on the picture to launch the movie) where you can see the thermal motions of 1.1  $\mu\text{m}$ -diameter fluorescent spheres mixed in water (left-hand side of the movie), and the trajectories I extracted from the same movie (right-hand side). Note that trajectories are measured individually (and I used different colors to display them). Also note that some particles are moving out of focus, which ends their tracking, and come back in focus, which starts a new trajectory (even if it is potentially the same particle).



## How do the data look like?

The output of multiple particle tracking are trajectories. Commonly, these trajectories are 2D, and are stored in an array that looks like that:

$x$	$y$	time $j$	id $i$
42.6914	81.6781	0.00000	0.00000
43.5721	82.8166	1.00000	0.00000
42.4250	81.9106	2.00000	0.00000
44.0587	83.6478	3.00000	0.00000
41.8534	83.0542	4.00000	0.00000
301.995	158.851	2.00000	1.00000
301.542	156.749	3.00000	1.00000
300.578	157.351	4.00000	1.00000
302.855	157.835	5.00000	1.00000
299.127	159.383	6.00000	1.00000
.	.	.	.
.	.	.	.
.	.	.	.

The 4th column is the trajectory identification number  $i = 0, 1, 2, \dots$ ; the 3rd is the time index  $j = 0, 1, 2, \dots$ ; the first two columns give the corresponding positions in the  $(x, y)$  imaging plane,

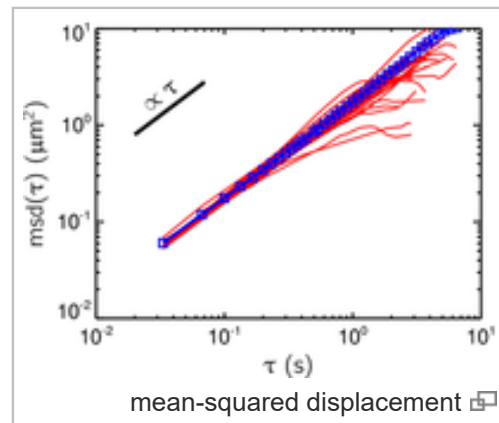
$$\mathbf{r}_{ij} = (x_{ij}, y_{ij})$$

of the trajectory labelled  $i$  at the time  $j$ . Eventually more information is extracted from the movie processing but it is not important for what we want to do here.

## An important quantity: the mean-squared displacement (msd)

The mobility of a colloidal tracer reflects the mechanical properties of the microenvironment it is embedded in. Analyzing data for a single particle, or groups of particles, allow us to quantify the mechanical properties of micro-scale environments. Since they are stochastic processes, the relevant physical observables we are interested in are *averages*. For reasons I will discuss in the next paragraph, an averaged quantity that is often calculated is called the mean-squared displacement. I will explain how to calculate this quantity in the next section [→](#), but I give here its generic formula:

$$\text{msd}(\tau) = \langle \Delta \mathbf{r}(\tau)^2 \rangle = \langle [\mathbf{r}(t+\tau) - \mathbf{r}(t)]^2 \rangle$$



where  $\mathbf{r}(t)$  is the position of the particle at time  $t$ , and  $\tau$  is the lag time between the two positions taken by the particle used to calculate the displacement  $\Delta \mathbf{r}(\tau) = \mathbf{r}(t+\tau) - \mathbf{r}(t)$ . The average  $\langle \dots \rangle$  designates a time-average over  $t$  and/or an ensemble-average over several trajectories. But at this point, it is important to understand that the mean-squared displacement can be calculated from a single trajectory by only performing a time average. If I calculate the msd vs.  $\tau$  for the Brownian particles in water I had tracked in the movie linked above, I find the results shown in the figure on the left. Again, I will give more details later on how I calculated this msd. Here I performed a time and ensemble average over all measured trajectories to obtain the blue points. Each red curve is a time-average calculation over a single trajectory. These red curves are affected by a greater statistical uncertainty than the blue curve, but all curves follow the same trends. The log-log plot shows in particular that in that case, the msd varies linearly with the lag time,  $\text{msd}(\tau) \propto \tau$ . This is a characteristic of the Brownian motion observed in a purely viscous (or Newtonian) fluid, such as water. We can push further this qualitative observation by using the formalism of microrheology.

## Microrheology

The mean-squared displacement characterizes the dynamics of the particles we are tracking: it shows the amplitude of the particle's motion at a characteristic time given by  $\tau$ . These dynamics are related to the mechanical properties of the medium in which the particles are moving. For example, particles in a continuum viscous material will have a mean-squared displacement growing linearly with the lag time, but the more viscous it is, the slower will be the rate of growth of  $\text{msd}(\tau)$  vs.  $\tau$ . In particular, in a viscous fluid we have:

$$\text{msd}(\tau) = 2dD\tau$$

where  $d$  is the dimensionality of the track ( $d=2$  here in 2-dimensions) and  $D = kT / (6\pi\eta a)$  is the diffusion coefficient. In this formula,  $k$  is the Boltzmann constant,  $T$  is the temperature,  $\eta$  is the viscosity of the fluid and  $a$  is the radius of the particles. From the blue curve of the plot shown above, we calculate

$$D = 0.43 \mu\text{m}^2 \cdot \text{s}^{-1} \Rightarrow \eta = 9.2 \times 10^{-4} \text{ Pa} \cdot \text{s}$$

which is the accepted value for the viscosity of water at 23°C, as compared to tabulated values obtained using different techniques. This type of calculations lies in the framework of (passive) microrheology, where the motion of Brownian particles can be used to measure the mechanical properties of the material they are embedded in. Generalizations of this relation exist and can be used to calculate the complex shear modulus spectrum from  $\text{msd}(\tau)$  to characterize viscoelastic behavior of the material in which particle tracking was performed. For the purpose of this tutorial, it is important to understand that individually calculated msd's (such as the red curves in this plot) can also be used to perform local microrheological measurements.

