

Homework Assignment 4

1. Compare two proteins of masses 27kDa and 54kDa. By how much does their diffusion coefficient in the same buffer solution differ?
2. For a confocal volume with width $w_{xy} = 250\text{nm}$ and aspect ratio $g = 6$, which concentration do you have to choose to achieve 1, 10 and 50 particles in the focus on average.
3. Estimate the retention time of particles with diffusion coefficients of $D = 10\mu\text{m}^2/\text{s}$, $D = 50\mu\text{m}^2/\text{s}$ and $D = 500\mu\text{m}^2/\text{s}$ inside the above described laser focus ($w_{xy} = 250\text{nm}$, $g = 6$) at a temperature of 37 C.
4. Look at the two FCS autocorrelation curves in Fig. 1 (No photophysical effects are incorporated!). Estimate for each graph how many particles N have been observed, how many different components there were, what their diffusion time τ_D was and possibly the number of particles in each species! Also estimate the diffusion coefficients D of the different particles ($w_{xy} = 250\text{nm}$). Assume equal quantum yields for all species.

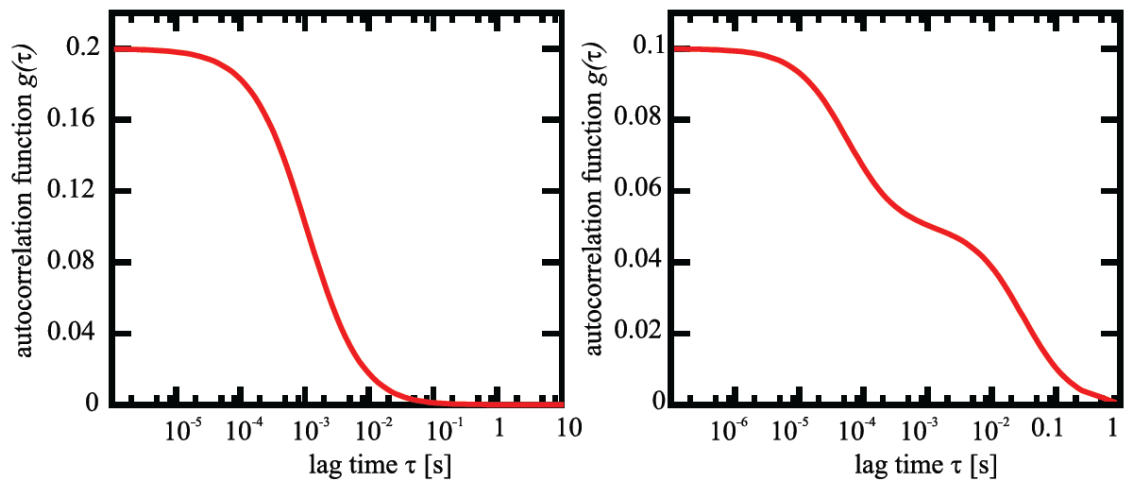


Fig 1. Example FCS autocorrelation curves. No photophysical effects are incorporated!