**Problem set #5 – diffusion and target search**

Please submit your answer before Thursday, January 16, 2025, 14:15 hour per e-mail to karsten.rippe@bioquant.uni-heidelberg.de and include [BPC2024] in the subject line.

1. The inert green fluorescent protein (GFP) has a free diffusion coefficient of *D* = 30 µm2 s-1 in the cell.

a) How long will it take a single GFP molecule with this value of *D* to diffuse in three dimensions the distance of

* the width of an E. coli cell (~1 µm)?
* the width of a "typical" nucleus of a human cell (~15 µm)?
* the length of an internodal cell in the alga Nitella (~1 cm)?
* the length of a spinal motor neuron that innervates a foot muscle in an adult human (~1 m)?

b) In which of the above cells is free diffusion not an efficient mechanism for the transport of macromolecules, and what could be alternative mechanisms in these cases?

c) Predict how a plot of the mean squared displacement (MSD) of GFP versus time for 0-10 seconds would look like in a human cell nucleus and compare it to a plot of MSD versus time for free diffusion of GFP.

2. The transcription factor p53 diffuses in vitro in three dimensions with *D*3 = 50 µm2 s-1 through a spherical volume of 15 µm diameter filled with water and a 24 base pairs long DNA fragment with its binding site.

a) Estimate the "search time" for p53 to bind the DNA under the above conditions according to the considerations given in the review by Berg & von Hippel (DOI: 10.1146/annurev.bb.14.060185.001023) on diffusion-controlled reactions.

b) Compare the “search time” you calculated in a) to the time it takes p53 to translocate from one end of the sphere to the other.

c) Name three different mechanisms by which the search time of p53 to find its target site in the cell could be reduced.

3. TFX can slide along long DNAs by one-dimensional diffusion with a diffusion constant *D*1 = 2·10-4 µm2 s-1. For unspecific binding to the DNA it dissociates with a kinetic rate constant of *k*off = 0.1 s-1 from the DNA.

a) On an average, the protein moves the length *l*slide along the DNA before it dissociates.

The value of *l*slide is given by 

Show how this expression for *l*slide can be derived.

b) Calculate *l*slide for the values of *D*1 and *k*off given above as well as the time it takes TFX to translocate a distance of *l*slide by diffusion in three dimensions with *D*3 = 50 µm2 s-1.

c) Discuss if one-dimensional diffusion along the DNA will speed up the search time of a transcription factor for finding its target DNA sequence with the parameters given above.