Supplemental Document

Biomedical Optics EXPRESS

Analysis of intracellular protein dynamics in living zebrafish embryos using light-sheet fluorescence single-molecule microscopy: supplement

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Supplement DOI: https://doi.org/10.6084/m9.figshare.16444320

Parent Article DOI: https://doi.org/10.1364/BOE.435103

Analysis of Intracellular Protein Dynamics in Living Zebrafish Embryos Using Light-Sheet Fluorescence Single-Molecule Microscopy: supplemental document

Supplement 1

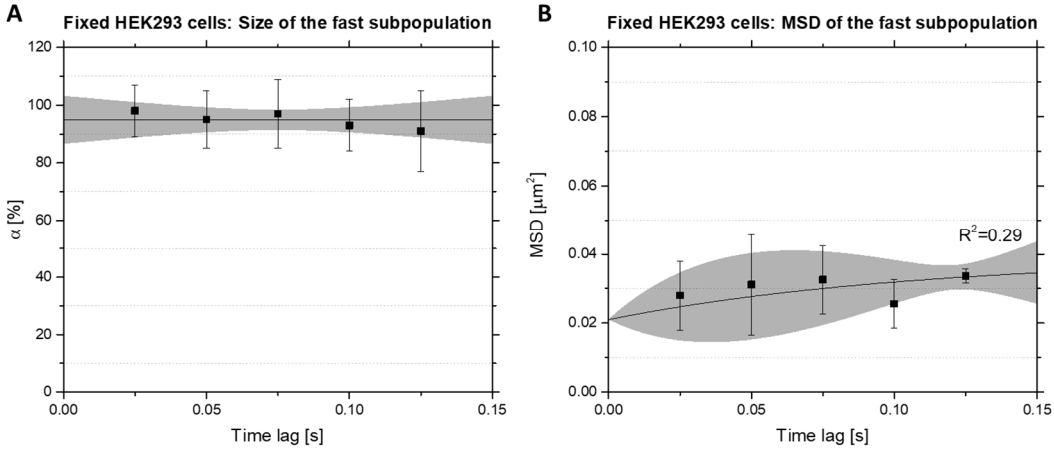

Fig. S1: Analysis of the mobility pattern of YFP-GR in fixed HEK293 cells. (A) Fast subpopulation fraction size fitted to a linear model with a fixed slope. Since 94.8 ± 1.3% of all the YFP-GR belong to one, fast-diffusing population, impact of the slow-diffusing fraction can in this case be neglected. (B) MSD values for the fast subpopulation, fitted using a confined diffusion model. The offset value of the measurement is 0.023 µm s^{-1}, and equals to $4 \cdot (dx)^2$, with the (dx) equaling 76 nm. The MSDs of this population are so close to the offset that the molecules can be considered immobile. In both graphs, the 95% confidence interval, standard deviation, and Pearson's correlation coefficients R^2 of the model fitness are shown.

Table S1: Results of the analysis of the mobility pattern of YFP-GR in fixed HEK293 cells

Parameter	Fixed HEK293 cells
$\boldsymbol{\alpha}$ [%]	94.8 ± 1.3
$\boldsymbol{D_0}$ fast subpopulation [µm^2 s^{-1}]	0.041 ± 0.038
$\boldsymbol{L}$ fast subpopulation [nm]	233 ± 151

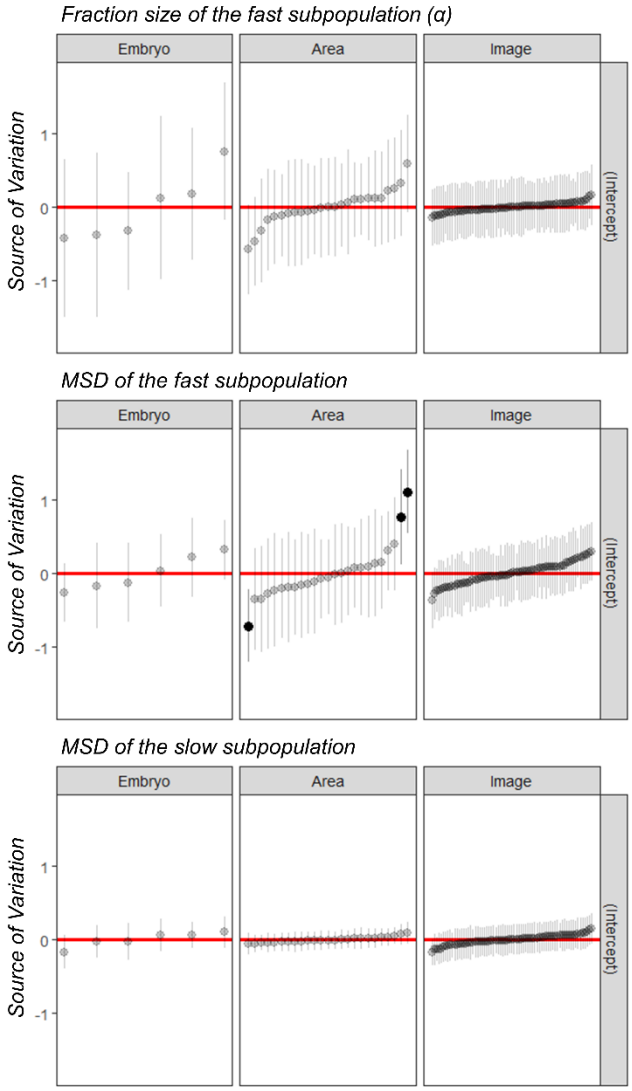

Fig. S2: Results of the mixed linear model analysis reveal potential sources of the data variability. To establish effect ranges, data were logit-transformed and presented on a logit scale. Caterpillar plots present the effect from random components as the deviation of the group intercepts from the global mean of the data. The effect range represents the deviation between different embryos, different areas within an embryo, and different images of the same area within an embryo, that are parts of the total measurement error in the mobile fraction size, α (A), MSD of the mobile fraction (B), and MSD of the immobile fraction (C). Red lines indicate the global mean of the data, while black dots signify significant deviations from the global mean among embryos, areas, and images. The data points are sorted from the ones most negatively deviating from the global average to the ones that deviate most positively.

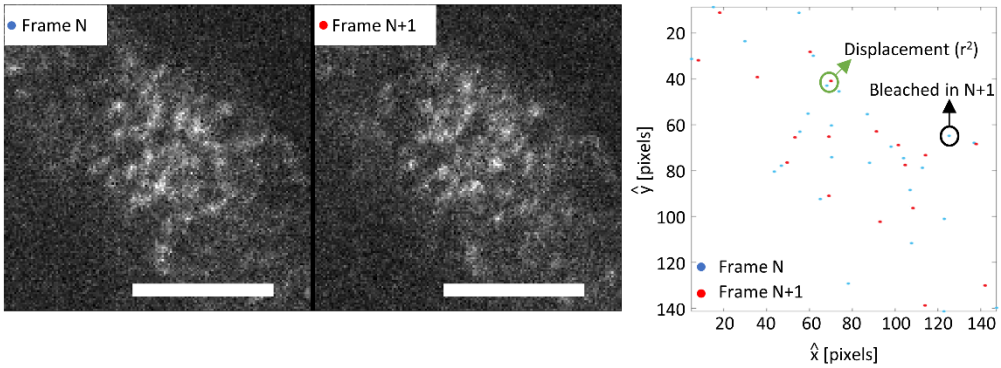

Fig. S3: Representative image of a displacement analysis based on a single image pair (Frames N and N+1). Not every molecule is positionally correlated due to the photobleaching. The average number of peaks per image here equals 23.0. This image pair is a part of a 3000-image movie. For the entire movie, the total number of displacements found in the 2999 image pairs equals to 35467.