

Oct 29, 2025

Monte Carlo simulation for FRAP procedures

DOI

<https://dx.doi.org/10.17504/protocols.io.n2bvjee5pgk5/v1>

Selçuk avuz¹, Bart Geverts², Martin E. van Royen¹, Adriaan B. Houtsmuller¹

¹Department of Pathology, Erasmus MC, Rotterdam, The Netherlands;

²Optical Imaging Center, Erasmus MC, Rotterdam, The Netherlands

Selçuk Yavuz



Selçuk Yavuz

Create & collaborate more with a free account

Edit and publish protocols, collaborate in communities, share insights through comments, and track progress with run records.

Create free account

OPEN  ACCESS



DOI: <https://dx.doi.org/10.17504/protocols.io.n2bvjee5pgk5/v1>

Protocol Citation: Selçuk avuz, Bart Geverts, Martin E. van Royen, Adriaan B. Houtsmuller 2025. Monte Carlo simulation for FRAP procedures . protocols.io <https://dx.doi.org/10.17504/protocols.io.n2bvjee5pgk5/v1>

License: This is an open access protocol distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited



Protocol status: Working

We use this protocol.

Created: October 28, 2025

Last Modified: October 29, 2025

Protocol Integer ID: 230963

Keywords: monte carlo simulation for frap procedure, quantitative analysis procedure for experimental frap curve, experimental frap curve, frap procedure, simulations of diffusion, monte carlo simulation, binding of protein, using monte carlo, based simulation, monte carlo, protein, diffusion

Disclaimer

- This work is based on previous work published by Geverts et al (DOI: [10.1007/978-1-4939-2080-8_7](https://doi.org/10.1007/978-1-4939-2080-8_7))

Abstract

This protocol describes briefly the quantitative analysis procedure for experimental FRAP curves by using Monte Carlo-based simulations of diffusion and binding of proteins.

- 1 Computer modelling used to generate FRAP curves for fitting was based on Monte Carlo simulation of diffusion and binding to immobile elements (representing chromatin binding) in an ellipsoidal volume (representing the nucleus). Bleaching simulation was based on experimentally derived three-dimensional laser intensity profiles, which determined the probability for each molecule to become bleached considering their 3D position relative to the laser beam. Diffusion was simulated at each new time step $t + \Delta t$ by deriving a new position $(x_{t+\Delta t}, y_{t+\Delta t}, z_{t+\Delta t})$ for all mobile molecules from their current position (x_t, y_t, z_t) by $x_{t+\Delta t} = x_t + G(r_1)$, $y_{t+\Delta t} = y_t + G(r_2)$, and $z_{t+\Delta t} = z_t + G(r_3)$, where r_i is a random number ($0 \leq r_i \leq 1$) chosen from a uniform distribution, and $G(r_i)$ is an inversed cumulative Gaussian distribution with $\mu = 0$ and $\sigma^2 = 2D\Delta t$, where D is the diffusion coefficient. Immobilization was derived from simple binding kinetics: $k_{on}/k_{off} = F_{imm} / (1 - F_{imm})$, where F_{imm} is the fraction of immobile molecules. The probability per unit time to be released from the immobile state was given by $P_{mobilise} = k_{off} = 1 / T_{imm}$, where T_{imm} is the characteristic time spent in immobile complexes expressed in unit time steps. The probability per unit time for each mobile particle to become immobilized (representing chromatin-binding) was defined as $P_{immobilise} = k_{on} = (k_{off} \cdot F_{imm}) / (1 - F_{imm})$, where $k_{off} = 1/T_{imm}$. Note that k_{on} and k_{off} in this model are effective rate constants with dimension s^{-1} .

In all simulations, the size of the ellipsoid was based on the size of the measured nuclei, and the region used in the measurements determined the size of the simulated bleach region. The laser intensity profile using the simulation of the bleaching step was derived from confocal images stacks of chemically fixed nuclei containing GFP that were exposed to a stationary laser beam at various intensities and varying exposure times. The unit time step Δt corresponded to the experimental sample rate of 100 ms.