SUBDIFFUSION SUPPORTS JOINING OF CORRECT ENDS DURING REPAIR OF DNA DOUBLE-STRAND BREAKS

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DNA double-strand breaks (DSB) are a major threat for the genomic integrity and thus cell survival. However, some repair pathways can mis-rejoin DNA ends from different breaks, creating chromosome aberrations. The mobility and initial distance of damaged chromatin regions in the nucleus may affect the probability of mis-repair. Repair processes can be made visible by tagging DNA repair proteins with the green fluorescent protein GFP, so that microscopic accumulations of the repair proteins ("foci") at the damage sites can be observed and analyzed "live" under a fluorescence microscope (cf. Fig. 1).

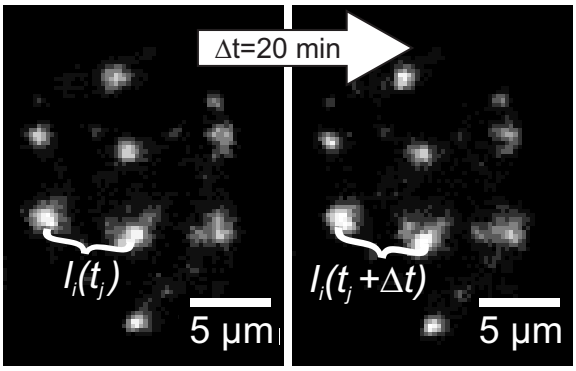

Figure 1: U2OS cell irradiated with carbon ions in a 5 x 5 µm^2 matrix pattern, visible under the fluorescence microscope by GFP-tagging of DNA repair protein MDC1 which accumulates at the radiation induced double-strand breaks ("foci"). The distance l_i between neighbouring foci is recorded at several time points t_j for analyzing the distance changes Δl_i over different time intervals Δt.

In this work, living U2OS osteosarcoma cells were irradiated in a 5x5 µm^2 matrix pattern with one carbon ion (43 MeV) per point or 32 protons (20 MeV) respectively at the ion microprobe SNAKE at the Munich 14 MV Tandem accelerator. Live-cell observation and distance tracking of the fluorescence-tagged damage response protein MDC1 was used to study the random-walk behaviour of chromatin domains containing ion-induced DSB. Our measurements indicate a subdiffusion-type random walk process with similar time dependence for isolated and clustered DSBs that were induced by proton or carbon ion micro-irradiation (cf. Fig. 2).

Subdiffusion is characterized by a Δt^{α} – dependence with $\alpha < 1$, whereas for normal diffusion $\alpha = 1$. We measure $\alpha = 0.50 \pm 0.06$.

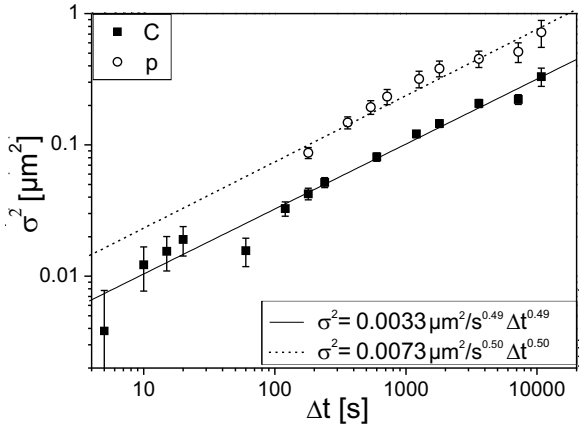

Figure 2: Double-logarithmic plot of the squared standard deviations σ^2 of the distance changes $\Delta l(\Delta t)$ between neighbouring MDC1 foci in the nuclei of cells irradiated with carbon ions (filled squares) and protons (open circles). The data are fitted with the power-law function $\sigma^2 = \Gamma \Delta t^{\alpha}$.

As compared to normal diffusion, subdiffusion enhances the probability that both ends of a DSB meet, therefore promoting high efficiency DNA repair. Furthermore, it also reduces their probability of long-range movements and thus lowers the probability of mis-rejoining and creation of chromosome aberrations. [1]

REFERENCES

[1] S.Girst et al., Sci. Rep. 3 (2013) 2511