Species conserved response at heterochromatic DNA damage *

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DNA double strand break (DSB) repair comprises several steps; besides different repair proteins also chromatin modifications are necessary and local as well as global chromatin decondensation takes place. Furthermore, recent findings in *Drosophila*, mouse, and baker's yeast show that DSBs within repetitive regions relocate to less repetitive chromatin [1-3]. Thus, relocation of DSBs might be a mechanism to prevent homology driven misrepair of repetitive sequences. Here we provide evidence that in the facultative heterochromatin of the human inactive X chromosome (Xi) decondensation occurs at DNA damage (Fig. 1 A and B) and DSBs are relocated to more open chromatin (Fig. 1 C, DSB marker 53BP1) [4]. Thus, DSB relocation is not limited to repetitive sequences but is a general process within repair of heterochromatic regions. Our results obtained in human cells indicate that decondensation at heterochromatic DSBs and their relocation is conserved from flies to mice and humans. In contrast to earlier findings [1-3], however, the DSB dependent chromatin modification of H2AX phosphorylation (γH2AX) shows not only relocation but additional H2AX phosphorylation takes place almost in the entire Xi area (Fig. 1 C, γH2AX signal). This increased γH2AX spreading is clearly LET (linear energy transfer) dependent as it is not seen upon carbon ion (not shown) but after calcium, titanium (not shown), or lead ion (Fig. 1 C) irradiation. The mechanism and functional relevance of this Xi wide γH2AX spreading will be analyzed in future studies.

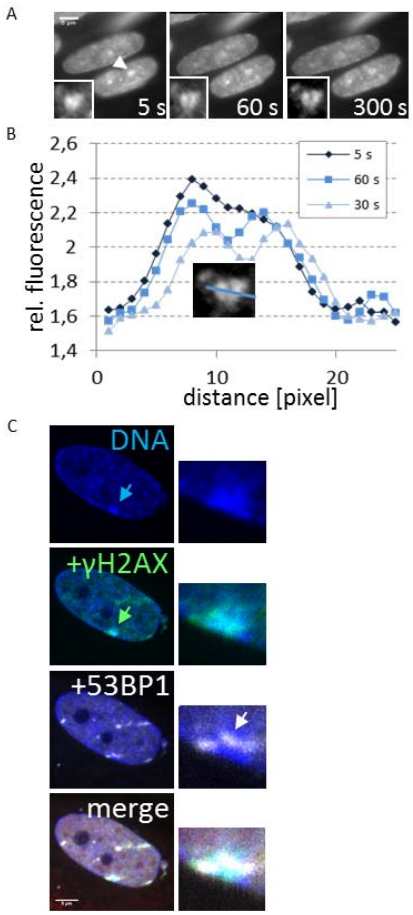

Figure 1: **(A)** Chromatin decondensation in the Xi chromosome of human cells. Real time DNA decondensation within Xi territories (arrow) identified by Hoechst 33342 DNA staining after irradiation with one gold ion at the GSI microprobe. Selected images of indicated time points are shown. The appearance of a darker area within the Xi (magnification in white box) represents the Xi decondensation at the ion hit. **(B)** The depletion of DNA staining intensities (i. e. occurrence of a darker area) over time at the site of an ion hit is exemplarily shown for intensity profiles along the line within the depicted Xi. **(C)** Relocation of heterochromatic DSBs is indicated by bending of 53BP1 (DSB marker) tracks around Xi 1 h after low angle lead ion (LET 13,500 keV/μm) irradiation (DNA signal: arrow indicates Xi; 53BP1 signal: arrow indicates bent 53BP1 track). Phosphorylation of H2AX occurs all over the ion hit Xi (arrow in γH2AX signal). Scale bars: 5 μm.

References

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