



Detection and study of DNA double-strand breaks repair after γ -irradiation

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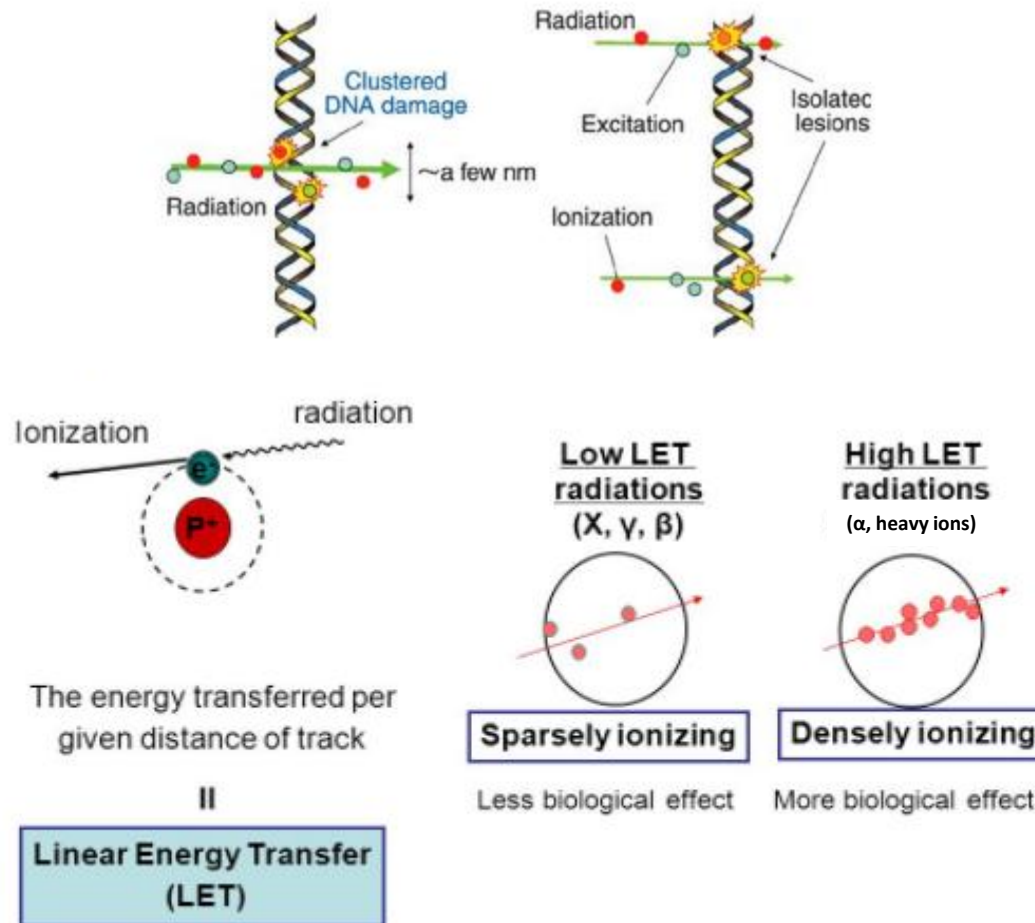
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Summary

- Introduction
- Aims and application
- Materials and methods
- Results
- Conclusions

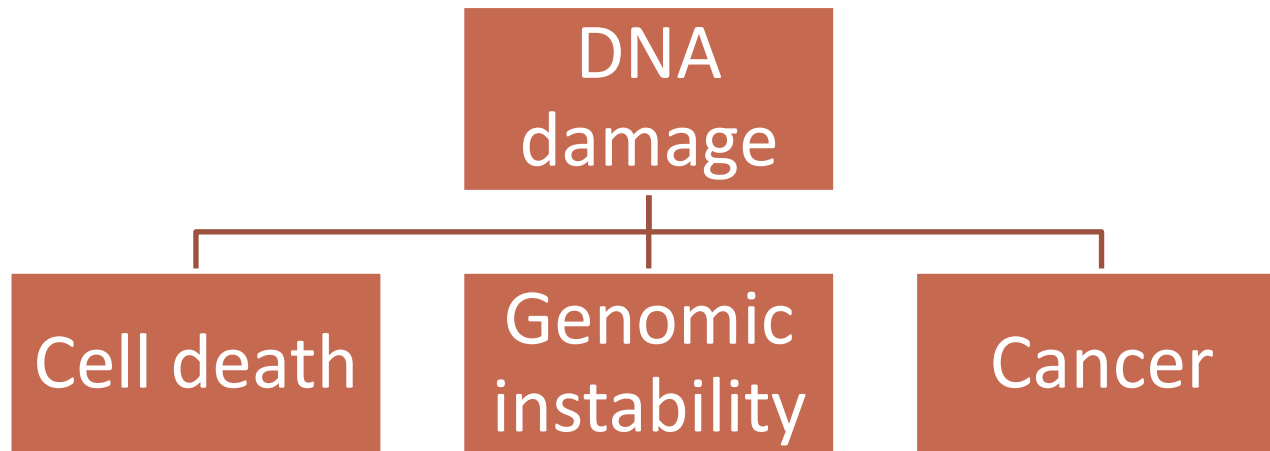
Differences between High- and Low-LET radiation

High-LET radiation causes dense ionization along its track, while Low-LET radiation produces ionizations sparsely along their tracks and almost homogeneously within a cell



DNA damage consequences

DNA double-strand breaks (**DSBs**) are extremely dangerous lesions which can lead to



DNA repair represents a cellular response to DNA damage for protecting our organisms from such deleterious consequences

Aim

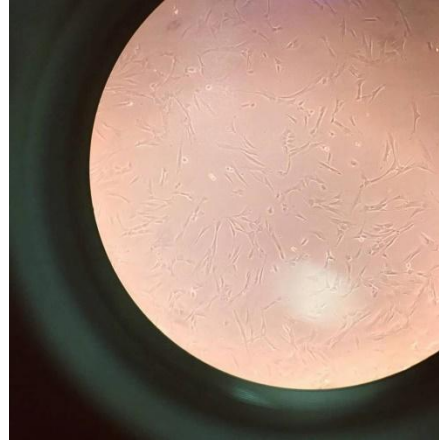
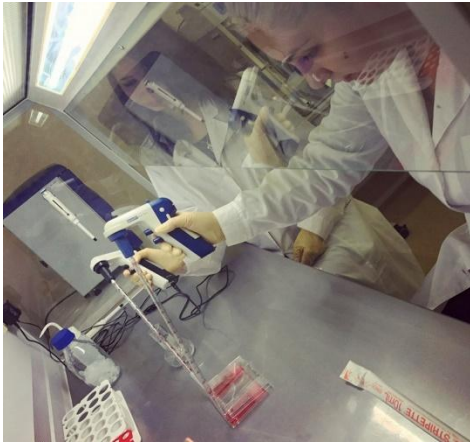
- Research of the dose dependence and repair kinetics of radiation-induced DNA DSBs on normal human fibroblasts exposed to γ -rays of ^{60}Co

Applications

- Diagnostics
- Cancer treatment
- Space radiobiology

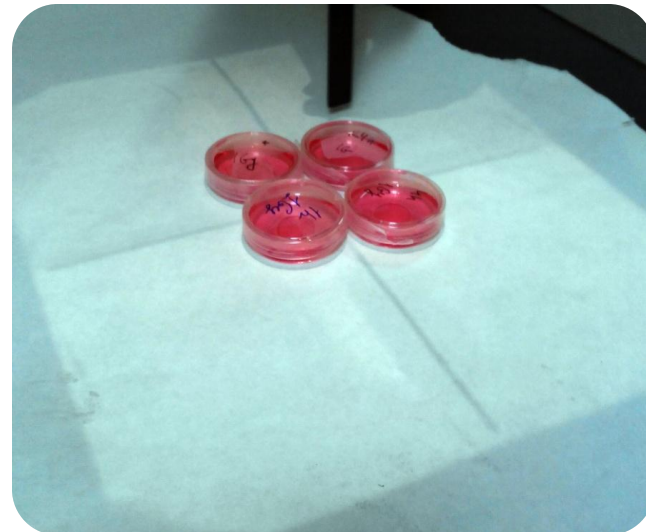
Materials and methods

Samples preparation



Irradiation with
 γ -rays of ^{60}Co
at Rocus-M facility

1Gy
3Gy
5Gy



Materials and methods

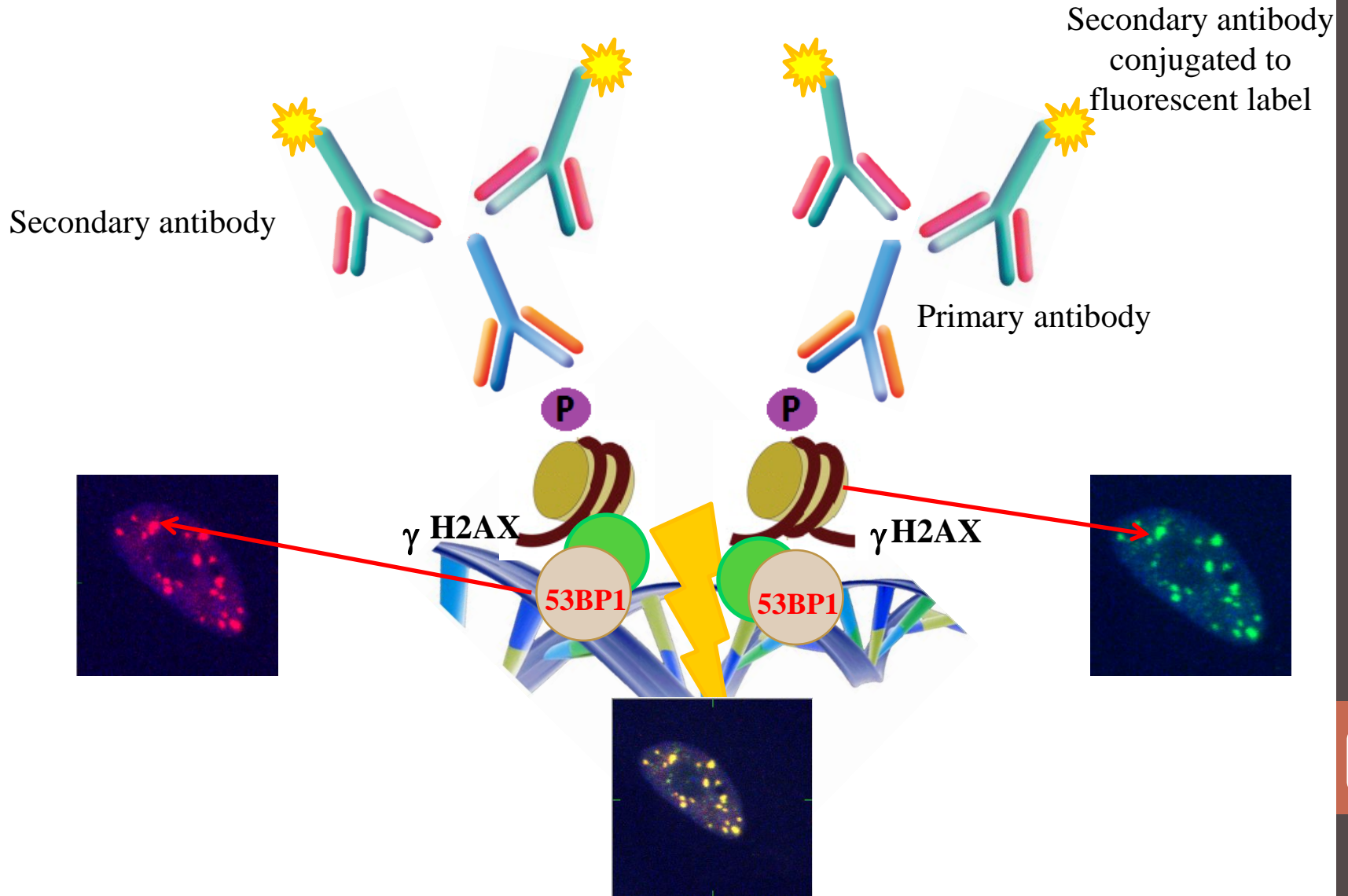
Immunostaining

- Fixation (1h, 4h and 24h)
- Permeabilization with Triton-X100 solution
- Incubation with primary ABs for gH2AX and 53BP1
- Incubation with secondary ABs conjugated with fluorescent markers

Microscopy



Visualization of induced DSBs *co-immunostaining of γ H2AX/53BP1 foci*



Materials and methods

Immunostaining

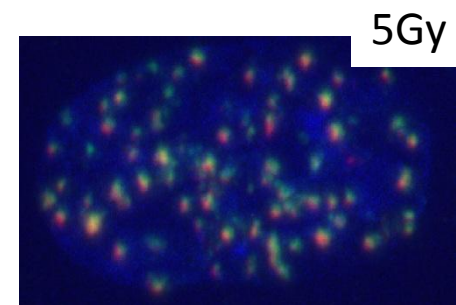
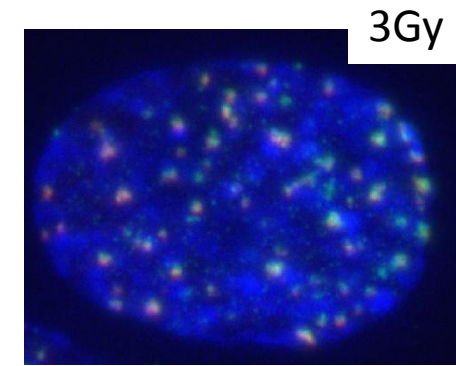
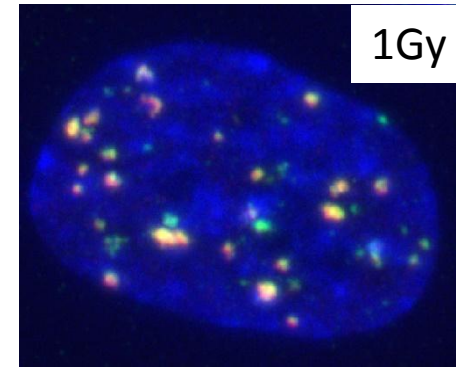
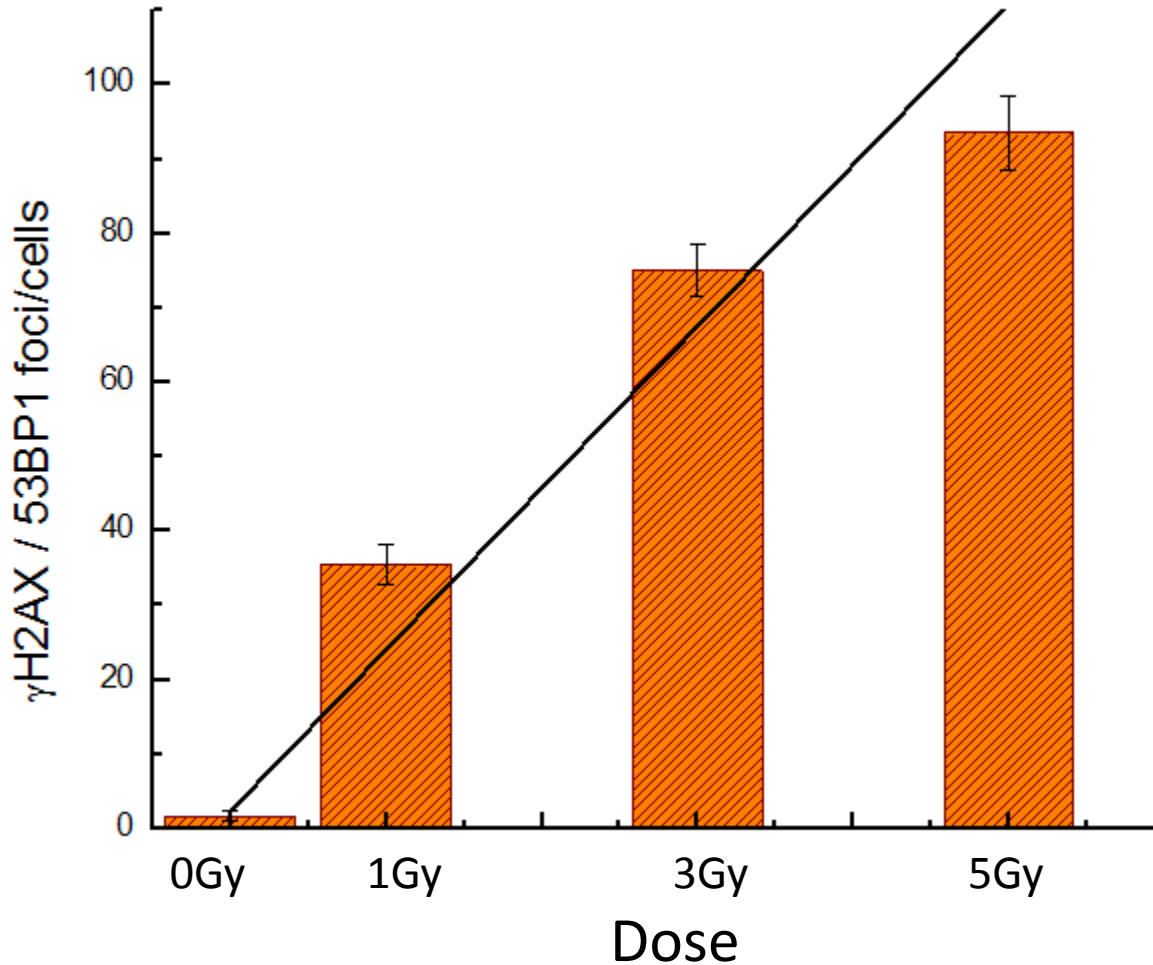
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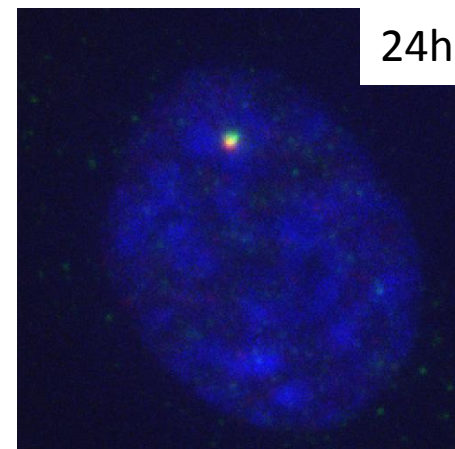
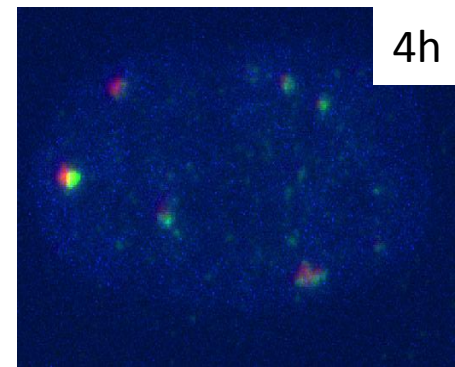
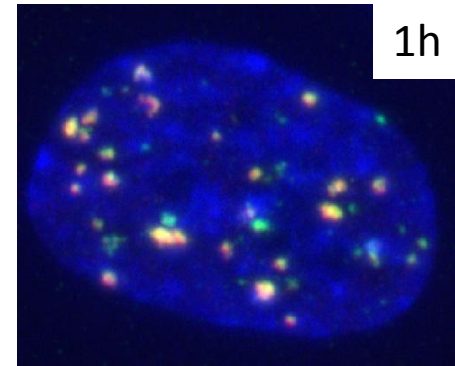
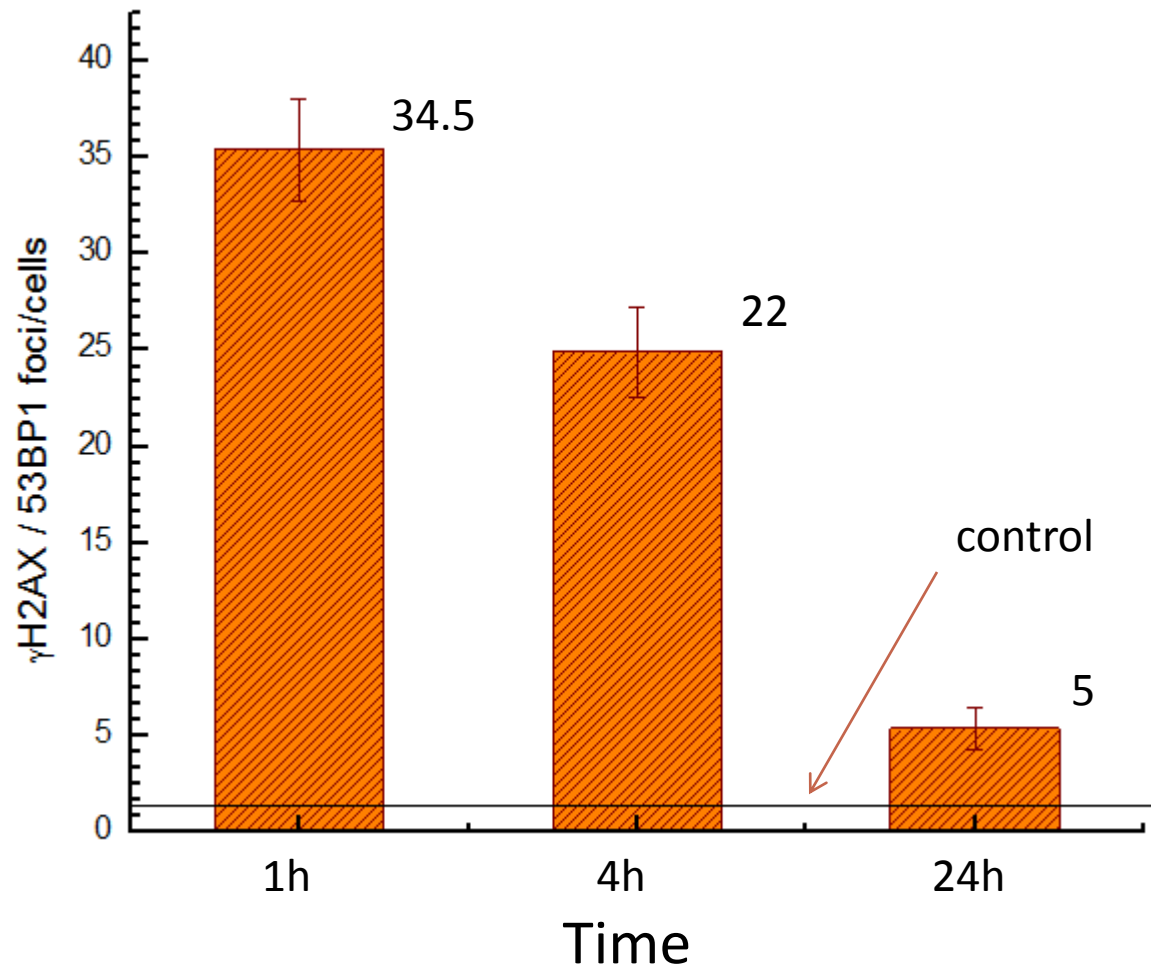


Image analysis with Acquarium software and ZenPro (Zeiss)

Dose response of the induction γ H2AX/53BP1 foci after 1 h post-irradiation with γ -rays



Elimination kinetics of γ H2AX/53BP1 foci after 1 Gy irradiation with γ -rays



Conclusions

- γ H2AX / 53BP1 foci increase with dose 1 h post-irradiation



Dose-dependence of DNA DSBs induction is close to linear

- Number of γ H2AX / 53BP1 foci decrease with time post irradiation



DNA DSBs are effectively repaired

Thank you for your attention !