

Evaluating the Potential of Radiosensitizers to Enhance the Effectiveness of Radiation Therapy

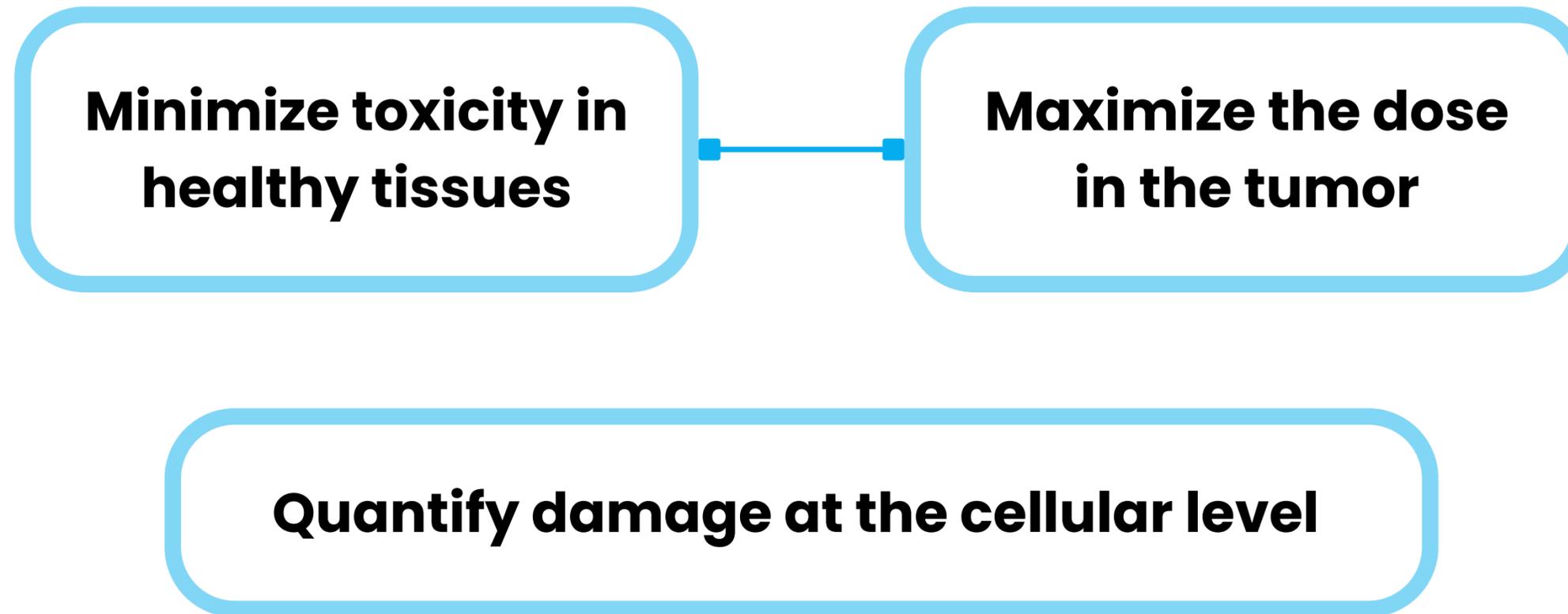
U.C. PIC2

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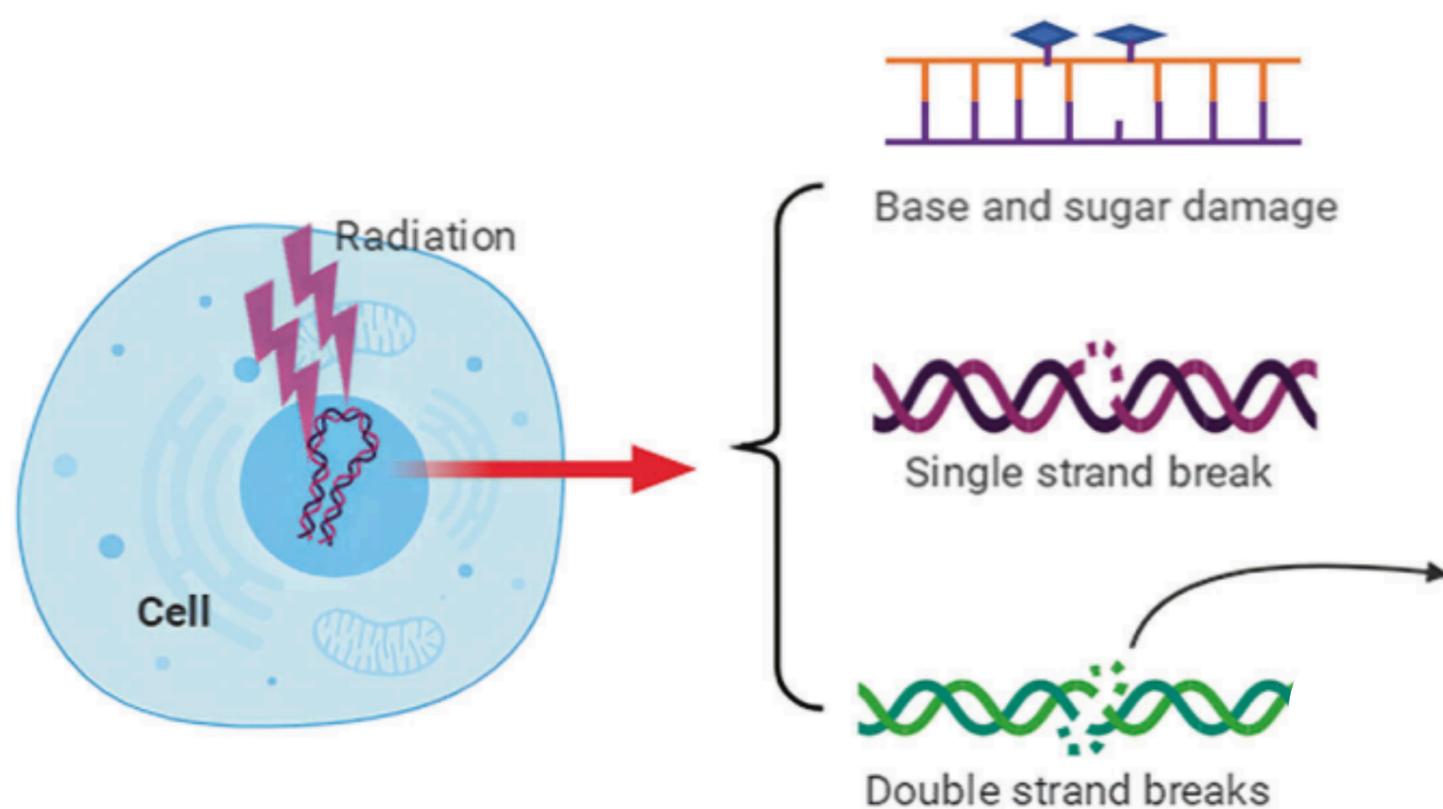
Central Goal



Biological Effects

DNA is the **most radiosensitive** cellular target

Double - strand breaks (DSBs) are the most critical damage



DNA damage biomarkers

- **γ -H2AX** \rightarrow DSB marker
- **53BP1** \rightarrow DNA repair marker

Nuclear foci

Confocal Microscopy

- **High spatial resolution**

- axial resolution between 400–800 nm
- lateral resolution ~ 200 nm

- **Z-stack** acquisition



- 3D reconstruction
- **Prevents foci overlap** seen in 2D
- **Improves quantification**

Image Analysis



Fiji: reference software for biological imaging

Plugin **FocAn:** Foci Analyser

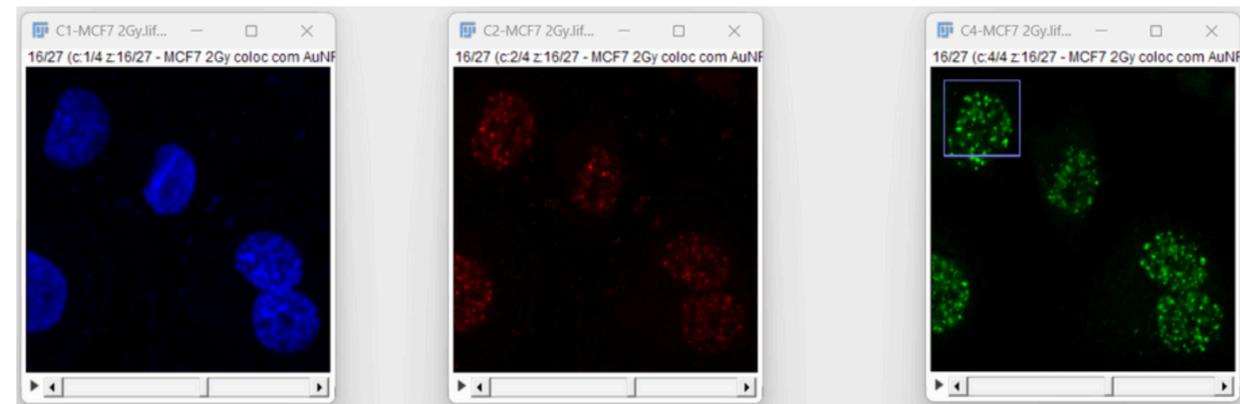
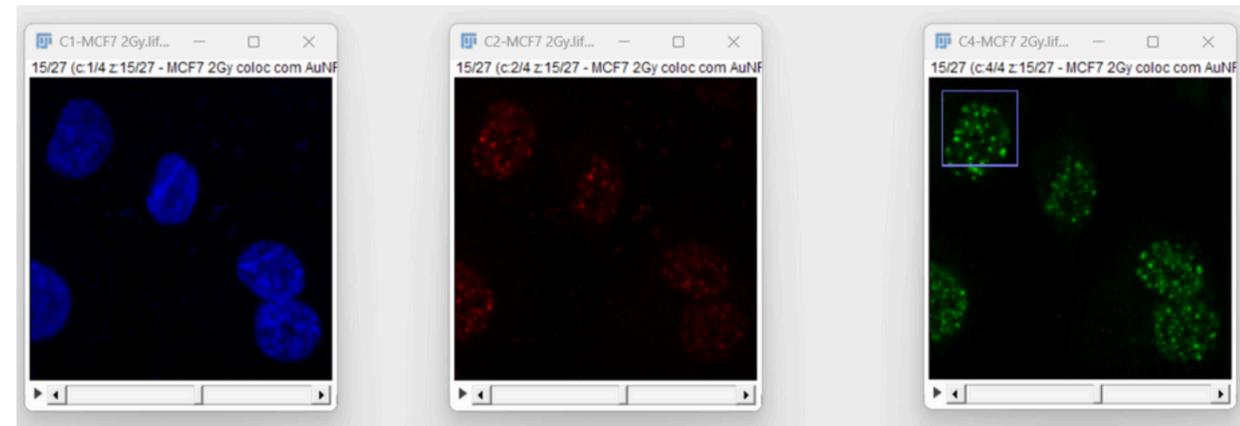
- analyzes multichannel
- automatically quantifies nuclear foci

FIJI: MCF-7 cells

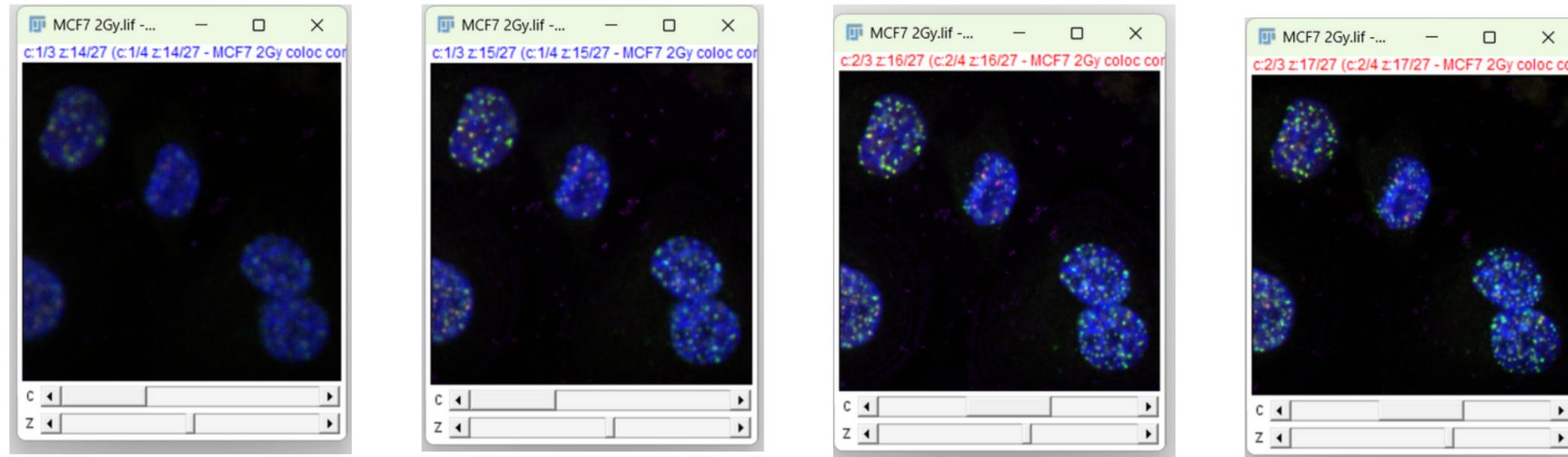
- **MCF-7** cell line (metastatic breast adenocarcinoma) incubated **with AuNPs** and irradiated with **2Gy X-rays**
- Multichannel imaging:
 - **Nuclei**: DAPI **blue** emission
 - **53BP1**: Texas **Red**-X emission
 - **γ -H2AX**: Alexa Fluor 488 emission (**green**)
- 13 to 16 stacks of 27 total stacks for this FOV



Z-depth influence on foci detection



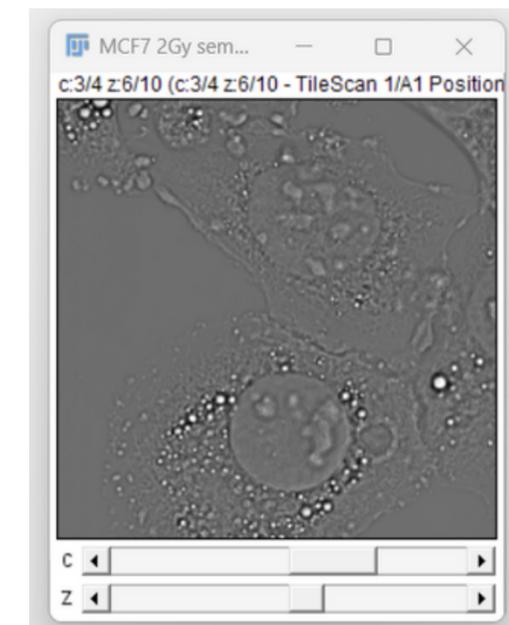
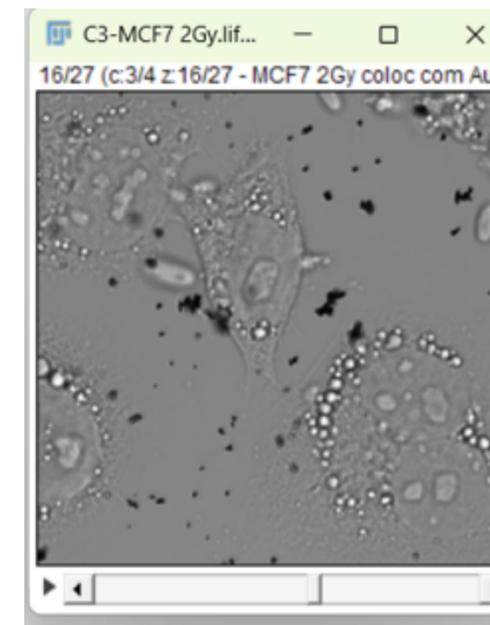
Colocalization



- Colocalization is visualized by channel merge, appearing as yellow regions
- 14 to 17 z-stacks of 27 showing intensity variation with depth

Presence of AuNPs

- Morphological channel confirms the presence of AuNPs
- AuNP-incubated sample (Left) exhibits black spots absent in control sample (Right)



With this analysis....

- Identification of energy deposition patterns
- Detailed study of how radiation, radiosensitizers, and DNA interact at the cellular level

Thank you