

Title : Impact of X-ray irradiation on a population of MCF-7 tumor cells at the single-cell level: a dynamic analysis using real-time microscopy

Keywords : videomicroscopy, radiobiology, in vitro, tracking

Abstract : Radiotherapy is a major component of cancer treatment, relying on the ability of ionizing radiation to induce cellular damage in order to limit or stop tumor cell proliferation. However, within a population exposed to the same radiation dose, individual cellular responses are highly heterogeneous and remain difficult to characterize using conventional approaches, which generally rely on population-averaged measurements.

This PhD work uses long-term live-cell videomicroscopy to investigate the response to irradiation at the single-cell level. Human MCF-7 breast cancer cells expressing cytoplasmic GFP were monitored for several days following X-ray irradiation. To analyze the large volume of data generated (hundreds of time-lapse videos and thousands of individual cells), an automated analysis pipeline named CellLineageTracking (CLT) was developed. CLT enables individual cell segmentation, tracking, and lineage tree reconstruction. Its performance was assessed using both standard tracking metrics and novel metrics specifically designed for this study.

Using CLT, the analysis of cell populations irradiated with doses up to 5 Gy and monitored over four days revealed marked heterogeneity in the response of cells exposed to the same dose. Three major cellular phenotypes were identified: proliferating cells, cells exhibiting a prolonged cell cycle, and cells undergoing persistent cell-cycle arrest. The relative proportions of these phenotypes varied with radiation dose.

They also displayed distinct morphological and dynamic characteristics: cells with prolonged or arrested cell cycles exhibited larger spreading areas and reduced motility. A fourth phenotype, corresponding to the daughter cells of cells with prolonged cell cycles, displayed an intermediate behavior: their cell area returned to values comparable to those of proliferating cells, whereas their motility remained higher than that of proliferating cells.

Another objective of this work was the investigation of the radiation-induced bystander effect, defined as the response of non-irradiated cells exposed to signals from neighboring irradiated cells. To this end, a co-culture strategy based on SNARF-SE fluorescent labeling was developed, enabling the simultaneous tracking of both cell populations using CLT.

Finally, the last part of this work investigates the use of platinum nanoparticles as radiosensitizing agents.