RADIATION-INDUCED CHROMOSOME DAMAGE ON MICROBEAM FACILITIES: DEVELOPMENT OF AN *IN SITU* PROTOCOL DESIGNED WITHIN THE *"BIOQUART"* PROJECT

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The biological effects of single charged particles cannot be simulated by *in vitro* conventional broad-beam exposures, due to the random Poisson distribution of particle tracks traversing the target cells. Charged-particle microbeam facilities were designed to target the nuclei or cytoplasm of single cells with a predefined number of particles and to analyze the induced damage on a cell-by-cell basis. In such a way, the radiation-induced cell damage can be directly correlated to type and energy of radiation and to the number of ions per cell. Within the *"BioQuaRT"* (Biologically weighted Quantities in RadioTherapy) Project, we developed an *in situ* protocol for the analysis of the unrepaired chromosome damage induced by charged particles irradiations at the PTB (Physikalisch-Technische Bundesanstalt, Braunshweig, Germany) microbeam facility. The development of a special *in situ* assay was required in this microbeam irradiation system because only a very limited number of cells (about 3000 cells/dish) could be seeded on the thin base made from BioFoil (25 μm thick) of the specific irradiation dishes designed at PTB. This method was developed on Chinese Hamster Ovary (CHO) cells, among the most commonly used cell lines in *in vitro* radiobiology experiments.

This protocol has the great advantage of allowing the simultaneous scoring of chromosome aberrations (CA) and micronuclei (MN) on the same irradiated sample. Although this method was developed for single-ion microbeam studies, it could be extended to other radiobiological applications requiring the use of *in situ* cytogenetic assays in case of restricted experimental conditions.

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