Application of the Fluorescence in situ Hybridization (FISH) Technique in Biological Dosimetry at CRCN-NE/CNEN

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Exposure to Ionizing Radiation (IR) can induce DNA damage, leading to structural chromosomal aberrations that serve as key biomarkers in biological dosimetry for dose-response assessments. While dicentric chromosome analysis is considered the gold standard for acute and recent exposures, its unstable nature causes it to decline over time with cell divisions, limiting its application in chronic or past exposures [1,2]. In this context, Fluorescence in situ Hybridization (FISH) using multicolor chromosome probes offers an effective alternative for retrospective biodosimetry by identifying stable chromosomal aberrations, such as translocations, which persist in circulating lymphocytes for decades [2-4]. This work aims to evaluate cellular responses by analyzing stable chromosomal aberrations in human peripheral blood lymphocytes exposed to 1 Gy of X-rays using the FISH assay. Peripheral blood was collected from a healthy, non-smoking donor with no prior history of radiation exposure. Irradiation was performed at the Metrology Service of the Centro Regional de Ciências Nucleares do Nordeste (CRCN/NE) with an absorbed dose of 1 Gy. Cell culture followed the International Atomic Energy Agency (IAEA) guidelines [1]. For FISH analysis, lymphocytes were fixed and hybridized with the XCP-mix probe set (MetaSystems), which specifically paints chromosome pairs 1, 2, and 4, followed by DAPI counterstaining [1,4]. Metaphase analysis was performed using a Leica DM-6000 fluorescence microscope equipped with Y3 and L5 filter sets. A preliminary evaluation of 3 slides yielded 85 viable metaphases compliant with ISO 20046:2019 criteria. Four stable chromosomal translocations involving chromosomes 1, 2 e 4 and DAPI-counterstained chromosomes were identified, corresponding to a frequency of approximately 4.71 translocations per 85 viable metaphases. These preliminary findings demonstrate the capability of the LDB/CRCN-NE to apply the FISH technique for stable aberration detection in retrospective dosimetry. Ongoing microscopic analysis will expand the sample size to establish statistically robust comparisons and integrate this dose point into the calibration curve under construction at CRCN-NE/CNEN.

References

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