GiC XXXIX WEB CONFERENCE 14 - 18 GIUGNO 2021
CONFERENZA NAZIONALE DI CITOMETRIA
AGGIORNAMENTI E INNOVAZIONI DELLA CITOMETRIA NELLE APPLICAZIONI CLINICHE E DI RICERCA

IONIZING RADIATIONS INDUCE MITOCHONDRIAL IMPAIRMENT IN NEUROBLASTOMA CELL LINES

Tortolici Flavia*, Incocciati Bernadette*, Aquilano Katia*, Lettieri-Barbato Daniele*, Giovanetti Anna°, Rufini Stefano*
*Department of Biology, University of Rome "Tor Vergata".
°ENEA, Department of Energy and Sustainable Economic, Rome, Italy

Aim. Ionizing radiations may induce in tumor cells unwanted responses leading not only to radio-resistance but also to an increase in the aggressiveness, that are often associated to metabolic changes. The aims of this work were to analyze in human neuroblastoma cell lines treated with X rays, the mitochondrial status and the effects induced by the secreted vesicles on the non-irradiated cells.
Materials and methods. Human neuroblastoma cell lines (SH-SY5Y) were irradiated with increasing doses of X-rays (1 to 10 Gy). The secreted vesicles were purified by the ultracentrifugation / filtration method and analyzed by flow cytometer. Then, cells irradiated with 10 Gy of X-rays after 24 hours were incubated with two fluorescent probes - i.e. MitoTracker™ Green and MitoTracker™ Red, capable of detecting respectively the mass of mitochondria and their transmembrane potential. The fluorescence intensity in the irradiated and non-irradiated cell populations was analyzed by flow cytometer and with confocal microscopy. The mass of mitochondria was further assessed through the determination of mtDNA by quantitative PCR analysis. These results were compared with the mitochondrial functionality by Seahorse Analyzer. This experimental approach was also applied to the non-irradiated neuroblastoma cells after their treatment with the vesicles secreted by the irradiated cells.
Results and discussion. Data obtained by flow cytometry indicate that X-rays treated neuroblastoma cells release an increased number of extracellular vesicles. Flow cytometry and confocal microscopy showed that X-rays induce a mitochondria impairment. Finally, the production of ATP and the consumption of oxygen decrease both in cells irradiated with X-rays and in those treated with extracellular vesicles.
Conclusions. The obtained results showed that X-rays induces a reduction in the involvement of mitochondria in cellular metabolism which leads to the establishment of a more glycolytic phenotype.