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Study of the reactive oxygen species induction in V79 cells after chemical treatment and irradiation

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The radiation can damage living organisms and their DNA, which is believed to be the critical target, in direct or indirect manner. The reactive oxygen species (ROS) are considered to be one of the most damaging agents causing the indirect effects of radiation (especially the hydroxyl radicals, but also peroxy radicals, superoxide radicals, etc.). It is speculated that the long-term elevated levels of ROS can cause unexpected irregularities in DNA and some hypothesize it could be a source of the inherited genomic instability. The work was conducted on the mammalian cell line of Chinese hamster (V79). The cells were exposed to gamma radiation of Co-60 at the Rokus-M unit of Dzhelepov Laboratory of Nuclear Problems. Applied doses were from 0.5 up to 10 Gy. As chemical inducers of ROS, the menadione and hydrogen peroxide were used. Changes in the levels of ROS after irradiation were measured with the use of general oxidative stress indicator (CMH2DCFDA) with fluorescent dye. This study is also aimed on the comparison of measurements in the cell suspension and in the adhered cells. Intensity of fluorescent light corresponding to the concentration of ROS in suspension of cells was measured on fluorometer Qubit 2.0 (Invitrogen). The fluorescent signal of cells adhered to the 96-well microplate was measured using the Multi-mode microplate reader Synergy H1 (Biotek) and the same fluorescent kit. Both approaches have their advantages and disadvantages which are discussed. There are still many uncertainties connected to the effects and persistency of the ROS in mammalian cells and a further research is essential.

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