

**Chromosomal Radiosensitivity, DNA repair, SNP and Apoptosis in Lymphocytes of Cervix Cancer Patients**

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There is considerable evidence that even after similar radiotherapy treatment, patients differ widely in normal tissue reaction. It is believed that the observed differences are related to intrinsic biological factors. Despite the great number of investigations of the relationship between the cellular radiosensitivity and reactions of patients to radiotherapy, there is presently no convincing proof that any of such biological factors alone can be used to predict side effects caused by radiotherapy. Considering the complexity of the mechanisms involved in the maintenance of the cell, tissue and organ function, it seems necessary to analyze the relationship between many different biological factors related to cellular sensitivity to ionizing radiation. The aim of the present study was to examine, in peripheral blood lymphocytes of cervix cancer patients collected before radiotherapy (1) chromosomal radiosensitivity, (2) kinetics of DNA repair, (3) SNP polymorphisms of genes involved in DNA repair and (4) apoptosis. The preliminary study included 30 patients. From each donor, peripheral blood was collected, irradiated in vitro with 2 Gy of gamma rays and the following endpoints were analyzed: Analysis of micronuclei (MN): The frequency of micronuclei, as a measure of chromosomal radiosensitivity, was determined with the micronucleus (MN) assay according to a standard protocol. Analysis of DNA repair: The kinetics of DNA repair was assessed by analyzing the formation and loss of gamma-H2AX foci. After 1 and 24 hours after irradiation, the cells were fixed, incubated with anti-phosphohistone H2AX antibody and analyzed by flow cytometry. Analysis of SNP polymorphism: DNA was isolated from the lymphocytes and RFLP-PCR followed by enzymatic digestion was performed according to the manufacturer's protocol. The XRCC1-Arg399Gln, XRCC3-IVS5-14.893 and OGG1-Ser326Cys polymorphisms were determined using the restriction enzymes HpaII, PvuII and Fnu4HI, respectively. Analysis of apoptosis: The frequency of spontaneous and radiation-induced apoptotic cells was determined after 1, 6 and 24 hours after irradiation by the annexin method and flow cytometry. The results of the present study have shown a substantial inter-patient variability of lymphocyte chromosomal radiosensitivity, kinetics of DNA repair, SNP polymorphisms and apoptosis. However, no signif-

icant relationship was found for any combination of these endpoints. Additional correlation analyses will be performed with the reaction of the healthy tissue of patients to radiotherapy, classified according to the EORTC/RTOG scale.