CELL TYPE DEPENDENT SIGNALING AND ITS EFFECT IN TISSUE REGULATION IN A HUMAN SKIN MODEL AFTER EXPOSURE TO LOW DOSES OF IONIZING RADIATION

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An early event after exposure to ionizing radiation is the induction of a broad cell signaling program which includes proliferation, differentiation, inflammation, and tissue remodeling. Previously, we have examined the X-ray radiation induced temporal response of an in vitro three dimensional (3D) human skin tissue model using microarray-based transcriptional profiling. Our data show that exposure to 10 cGy of low linear energy transfer (LET) X-rays is sufficient to affect gene transcription. Cell type specific analysis showed significant changes in gene expression with the levels of > 1400 genes altered in the dermis and > 400 genes regulated in the epidermis. The two layers rarely exhibited overlapping responses at the mRNA level. Quantitative reverse transcription polymerase chain reaction (qRT-PCR) measurements validated the microarray data in both regulation direction and value. Key pathways identified relate to cell cycle regulation, immune responses, hypoxia, reactive oxygen signaling, and DNA damage repair. Individual genes involved in extracellular matrix (ECM) remodeling were found to be strongly up-regulated. A fundamental question is whether the relative contribution of different signaling pathways remains the same as a function of radiation quality.

To address the question of low dose signaling pathway activation by high LET radiation we have examined the temporal responses of the 3D skin model post exposure to 600 MeV/u Fe, 500 MeV/u Ti, 400 MeV/u Si, 300 MeV/u Ne, 250 and 500 MeV/u O ions. All samples were exposed to constant ion fluences (3.6 x 10⁴ and 1.1 x 10⁵ particles/cm²). By following this irradiation protocol, we can focus on the delta ray contribution to the induced signaling. Based on the results measured in low LET exposed samples, we concentrated our high LET analysis on the same pathways. Proliferation, differentiation, wound healing, and tissue remodeling as well as the integrity of the barrier function were studied in mRNA, protein and histology samples. While the expression of the differentiation markers keratin 10 and filaggrin were mainly unaffected with increasing dose and time in ion exposed samples, the thickness of the differentiated layer increased. The analysis of time matching histological samples stained for incorporation of EdU as a proliferation marker did not identify proliferation as the cause for the increase in thickness. The outmost cell layers at the skin-air interface comprise the the skin barrier. Disruption of the skin barrier allows the entry of pathogenic germs which can cause severe infected wounds. Exposure to heavy ions does not appear to affect skin barrier function. In previous experiments with X-ray exposed samples we found genes regulated involved in wound healing pathways. For Fe and Ne exposed samples a qRT-PCR based array was used to analyze relevant genes for skin wound healing. The gene amplification data showed that all exposed samples regulated genes related to a wound healing pathway but different genes were regulated at the various time points. All data obtained will feed into a model development for prediction of early cell signaling events which might lead into long-term health consequences.