Characterization of the effects of gamma and proton radiation on immune cells using genetic and metabolomics approaches

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ABSTRACT
The objectives of this recently initiated study are to investigate acute and persistent effects of proton radiation on immune cell subsets and function. The role(s) for p38 MAP kinase in such radiation responses is being investigated using a genetic approach where an engineered mouse line has had one wt p38α gene replaced with a dominant-negative mutant (p38α+/DN). T cells are one of the most radiosensitive cell types in vivo, and radiation is known to impact CD4 T cell function long term. T cells are normally activated by antigen, which triggers differentiation to specific subsets involving various cytokines. In addition, T cells have a well-characterized cellular program upon activation by T-cell receptor (TCR) signaling that is reflected by major changes in metabolism. Metabolomics, which measures overall small molecules (<1 kD) in a sample, is ideal to detect many of these IR responses, and represents a global approach to assess changes in metabolism.

We compared the sensitivity of subsets of immune cells to ionizing radiation with wild type mice (wt) and p38α+/DN model. Splenocytes were isolated from mice at one and two weeks after radiation. The subsets of splenic lymphocytes were determined by FACS analysis. We observed that the percentage of CD4-, CD8- population in T cells increased after -radiation exposure, which suggest this subpopulation of T cells are more resistant to radiation. Our data showed that in p38α+/KI mice the increase of CD4/CD8 ratio was greater than wt. Further studies are needed to elucidate the role of p38 in specific subset responses. We also evaluated the effect of radiation on T cell activation. In wt mice the isolated T cells showed significantly compromised competence in responding to TCR mediated activation after radiation. In addition to measuring classical endpoints such as cell proliferation and cytokine production for evaluating T cell activation, we applied our metabolomics approach. We observed a variety of metabolic changes during activation in unirradiated T cells, and observed changes in these profiles with T cells from irradiated mice. Our results showed that T cell activation is remarkably compromised with 1 Gy proton radiation. Interestingly the effects on T cells’ activation and metabolite changes are dose-rate dependent. Taken together. Our results indicate that radiation remarkably affects T cell activation and proton exposure showed distinctive changes comparing to low LET radiation.