Molecular Mechanism Underlying Cellular Adaptive Response to Low Dose Radiation

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The goal of this research is to identify the molecular mechanisms by which cells adapt to low dose radiation exposure. Previously we have shown a radiation dependent increase of Calmodulin (CaM) in RAW 264.7 macrophages (RAW). Therefore we hypothesize that CaM and associated signaling complexes are sensors of low-dose radiation, resulting in alterations in energy metabolism and gene expression. The ultimate experimental goal for the investigation of this hypothesis is to obtain detailed proteomic time course data for cells exhibiting an adaptive response. In preparation, an Accurate Mass Tag (AMT) database has been created for the RAW and CaM over expressing RAW 264.7 (RAWC) cells. The AMT database not only provides a tool for future analysis of time course data[6], but currently allows the semi-quantitative analysis of protein abundance between the RAW and RAWC cell lines. Changes in protein levels due to the two fold increase of CaM observed in the RAWC cells[7] offer an understanding of the potential role of CaM in the radio-adaptive response.

This global proteomic analysis identified 4301 proteins with a notable change in abundance for greater than 100 proteins. Examination of proteins that are both increased and decreased in RAWC cells suggest a link between CaM and proteins reported to be involved in the adaptive response. Specifically two proteins that show an increase in the RAWC cell line are AHNAK, a nuclear phosphoprotein and the growth factor receptor-bound protein 2 (GRB2). Both have a potential link to the adaptive response mechanism via protein kinase C (PKC) or the epidermal growth factor receptor (EGFR), which have been implicated in the radiation-adaptive response[5]. Importantly, AHNAK has been shown to translocate to the plasma membrane in a PMA stimulated cell line[3]. In the plasma membrane it is proposed to function as a scaffold protein facilitating the activation of phosphoinositide-specific phospholipase C-γ (PLC-γ) and PKC-α[3], thus allowing for a rapid and efficient signal transduction process. GRB2 is known to be activated by the epidermal growth factor receptor (EGFR)[8], but it also interacts with PLC-γ[1,2]. Therefore it is possible that a signaling complex may exist that includes CaM and plays a role in the adaptive response.

Additional proteins of interest include increased abundance of Sumo-1, which has been shown to modulate tumor suppressor P53[4], a decrease in the pro-inflammatory cytokine Interleukin-16 and a decrease in the transcription factor NFAT5, which has a dependence on the calcium-calmodulin-dependent phosphatase calcineurin.

The combination of changes in protein abundance due to increased CaM expression, recently acquired proteomic analysis of CaM binding partners and known protein responses to radiation from the literature will allow for a more targeted investigation of the mechanism involved in the radio-adaptive response.

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