

Mouse Strain-Dependent Variations in Sensitivity to Induction of Gamma-H2AX Foci after Continuous Low Dose-Rate Irradiation: The *Atm*<sup>-/-</sup> vs *Atm*<sup>+/+</sup> genotypes on Balb/c, 129S6, C57BL/6J, and A/J inbred strains.

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We have recently developed a ‘low dose rate’ gamma-H2AX assay that is sufficiently sensitive to distinguish differences in response for cells from normal *Atm*<sup>+/+</sup> (mouse) or *ATM*<sup>+/+</sup> (human) and the phenotypes associated with the corresponding heterozygous genotypes(1, 2). The assay is also capable of distinguishing mild hypersensitivities for cells from an appreciable proportion of apparently normal individuals(3). We used this assay to determine whether the genetic background of four commonly used inbred mouse strains showed any differences in radiosensitivity for this assay, and second, whether the *Atm*<sup>-/-</sup> genotype on these same backgrounds resulted in a uniformly increased radiosensitivity that might further add to or reduce the sensitivity of the mice on these different backgrounds. Fibroblast cultures were established from ear clips from three separate mice of each mouse strain and cells in microscope chamber flasks were allowed to grow to a confluent monolayer and the medium was replaced with isoleucine deficient medium. After two days the cells were in a G0/G1 state, and the cultures were then irradiated with Cs-137 gamma-rays at a dose rate of approximately 8.6cGy/h for 24 hours. After irradiation the cells were fixed and immunostained to detect gamma-H2AX foci. These foci were detected in the fluorescence microscope using an Metamorph system to capture images for analysis. Our preliminary experiments indicate that for the C57BL/6J, A/J, and 129S6 mouse strains there was a significant difference between the mean foci per cell for the *Atm*<sup>+/+</sup> vs the *Atm*<sup>-/-</sup> genotypes but no significant difference for the two *Atm* genotypes on the Balb/c strain background. The number of foci per cell for cells from the Balb/c *Atm*<sup>+/+</sup> mice was significantly higher than that for cells from the other *Atm*<sup>+/+</sup> strains. This is not altogether surprising if the *Prkdc* defect, which results in a known DNA dsb rejoining deficiency in the Balb/c mouse(4) is associated with the same epistasis group as *Atm* .

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Reference List

1. T. A. Kato, H. Nagasawa, M. M. Weil, J. B. Little, J. S. Bedford, *Radiat. Res.* **166**, 443 (2006)
2. T. A. Kato *et al.*, *Radiat. Res.* **166**, 47 (2006).
3. T. A. Kato *et al.*, *DNA Repair (Amst)* **6**, 818 (2007).
4. R. Okayasu *et al.*, *Cancer Res.* **60**, 4342 (2000).